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(54) Title: CDR-GRAFTED ANTI-TISSUE FACTOR A (57) Abstract	NTBO	DIE	S AND METHODS OF USE THEREOF		
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monoclonal antibodies against tissue factor but have reduce and are thus useful in the treatment and prophylaxis of h CDR-grafted antibodies and pharmaceutical compositions	numan t	thron	nbotic disease. The invention also provides	are potent anticoagular methods of making the	
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# CDR-GRAFTED ANTI-TISSUE FACTOR ANTIBODIES AND METHODS OF USE THEREOF

#### FIELD OF THE INVENTION

1

Monoclonal antibodies capable of inhibiting tissue factor (TF) are useful as anticoagulants.

Conventional rodent monoclonal antibodies, however, have limited use in human therapeutic and diagnostic applications due to immunogenicity and short serum half-life. The present invention provides CDR-grafted monoclonal antibodies against TF that retain the high binding affinity of rodent antibodies but have reduced immunogenicity. The present humanized antibodies are potent anticoagulants and are thus useful in the treatment and prophylaxis of human thrombotic disease. The invention also provides methods of making the CDR-grafted antibodies and pharmaceutical compositions for the attenuation or prevention of coagulation.

## 20 BACKGROUND OF THE INVENTION

The coagulation of blood involves a cascading series of reactions leading to the formation of fibrin. The coagulation cascade consists of two overlapping pathways, both of which are required for hemostasis. The intrinsic pathway comprises protein factors present in circulating blood, while the extrinsic pathway requires tissue factor, which is expressed on the cell surface of a variety of tissues in response to vascular injury. Davie et al., 1991, Biochemistry 30:10363.

Agents that interfere with the coagulation cascade, such

-2-

as heparin and coumarin derivatives, have well-known therapeutic uses in the prophylaxis of venous thrombosis. Goodman and Gilman, eds., 1980, The Pharmacological Basis of Therapeutics, MacMillan Publishing Co., Inc., New York.

Tissue factor (TF) has been investigated as a target for anticoagulant therapy. TF is a membrane glycoprotein that functions as a receptor for factor VII and VIIa and thereby initiates the extrinsic pathway of the coagulation cascade in response to vascular injury.

In addition to its role in the maintenance of hemostasis by initiation of blood clotting, TF has been implicated in pathogenic conditions. Specifically, the synthesis and cell surface expression of TF has been implicated in vascular disease (Wilcox et al., 1989, Proc. Natl. Acad.

Sci. 86:2839) and gram-negative septic shock (Warr et al., 1990, Blood 75:1481).

Ruf et al. (1991, Thrombosis and Haemostasis 66:529) characterized the anticoagulant potential of murine monoclonal antibodies against human TF. The inhibition of TF function by most of the monoclonal antibodies that were assessed was dependent upon the dissociation of the TF/VIIa complex that is rapidly formed when TF contacts plasma. Such antibodies were thus relatively slow inhibitors of TF in plasma. One monoclonal antibody, TF8-5G9, was capable of inhibiting the TF/VIIa complex without dissociation of the complex, thus providing an immediate anticoagulant effect in plasma. Ruf et al. suggest that mechanisms that inactivate the TF/VIIa complex, rather than prevent its formation, may provide strategies for interruption of coagulation in vivo.

The therapeutic use of monoclonal antibodies

l against TF is limited in that currently available
monoclonals are of rodent origin. The use of rodent
antibodies in human therapy presents numerous problems,
the most significant of which is immunogenicity.

Repeated doses of rodent monoclonal antibodies have been found to elicit an anti-immunoglobulin response termed human anti-mouse antibody (HAMA), which can result in immune complex disease and/or neutralization of the therapeutic antibody. See, e.g., Jaffers et al. (1986)

10 <u>Transplantation 41</u>:572. While the use of human monoclonal antibodies would address this limitation, it has proven difficult to generate large amounts of human monoclonal antibodies by conventional hybridoma technology.

Recombinant technology has been used in an 15 effort to construct "humanized" antibodies that maintain the high binding affinity of rodent monoclonal antibodies but exhibit reduced immunogenicity in humans. Chimeric antibodies have been produced in which the 20 variable (V) region of a mouse antibody is combined with the constant (C) region of a human antibody in an effort to maintain the specificity and affinity of the rodent antibody but reduce the amount of protein that is nonhuman and thus immunogenic. While the immune response 25 to chimeric antibodies is generally reduced relative to the corresponding rodent antibody, the immune response cannot be completely eliminated, because the mouse V region is capable of eliciting an immune response. Lobuglio et al. (1989) Proc. Natl. Acad. Sci. 86:4220; 30 Jaffers et al. (1986) Transplantation 41:572.

In a recent approach to reducing

- l immunogenicity of rodent antibodies, only the rodent complementarity determining regions (CDRs), rather than the entire V domain, are transplanted to a human antibody. Such humanized antibodies are known as CDR-
- 5 grafted antibodies. CDRs are regions of hypervariability in the V regions that are flanked by relatively conserved regions known as framework (FR) regions. Each V domain contains three CDRs flanked by four FRs. The CDRs fold to form the antigen binding
- 10 site of the antibody, while the FRs support the structural conformations of the V domains. Thus by transplanting the rodent CDRs to a human antibody, the antigen binding domain can theoretically also be transferred. Owens et al. (1994) J. Immunol. Methods
- 15 <u>168</u>:149 and Winter et al. (1993) <u>Immunology Today 14</u>:243 review the development of CDR-grafted antibodies.

Orlandi et al. (1989) Proc. Natl. Acad. Sci.

USA 86:3833 constructed a humanized antibody against the relatively simple hapten nitrophenacetyl (NP). The CDRgrafted antibody contained mouse CDRs and human FRs, and exhibited NP binding activity similar to the native mouse antibody. However, the construction of CDRgrafted antibodies recognizing more complex antigens has resulted in antibodies having binding activity

- 25 significantly lower than the native rodent antibodies.

  In numerous cases it has been demonstrated that the mere introduction of rodent CDRs into a human antibody background is insufficient to maintain full binding activity, perhaps due to distortion of the CDR
- 30 conformation by the human FR.

For example, Gorman et al. (1991) Proc. Natl.

1 Acad. Sci. 88:4181 compared two humanized antibodies against human CD4 and observed considerably different

avidies depending upon the particular human framework region of the humanized antibody. Co et al. (1991)

- 5 Proc. Natl. Acad. Sci. USA 88:2869 required a refined computer model of the murine antibody of interest in order to identify critical amino acids to be considered in the design of a humanized antibody. Kettleborough et al. (1991) Protein Engineering 4:773 report the
- 10 influence of particular FR residues of a CDR-grafted antibody on antigen binding, and propose that the residues may directly interact with antigen, or may alter the conformation of the CDR loops. Similarly, Singer et al. (1993) J. Immunol. 150:2844 report that
- optimal humanization of an anti-CD18 murine monoclonal antibody is dependent upon the ability of the selected FR to support the CDR in the appropriate antigen binding conformation. Accordingly, recreation of the antigenbinding site requires consideration of the potential
- 20 intrachain interactions between the FR and CDR, and manipulation of amino acid residues of the FR that maintain contacts with the loops formed by the CDRs. While general theoretical guidelines have been proposed for the design of humanized antibodies (see, e.q., Owens et al.), in all cases the procedure must be tailored and
- 25 et al.), in all cases the procedure must be tailored and optimized for the particular rodent antibody of interest.

There is a need in the art for humanized antibodies with reduced immunogenicity and comparable 30 binding affinity relative to the parent rodent antibody for various therapeutic applications. In particular,

-6-

there is a need for a humanized antibody against human l tissue factor having anticoagulant activity and useful in the treatment and prevention of thrombotic disease.

#### SUMMARY OF THE INVENTION

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The present invention is directed to CDR-grafted antibodies capable of inhibiting human tissue factor wherein the CDRs are derived from a non-human monoclonal antibody against tissue factor and the FR and 10 constant (C) regions are derived from one or more human antibodies. In a preferred embodiment, the murine monoclonal antibody is TF8-5G9.

In another embodiment, the present invention provides a method of producing a CDR-grafted antibody
15 capable of inhibiting human tissue factor which method comprises constructing one or more expression vectors containing nucleic acids encoding CDR-grafted antibody heavy and light chains, transfecting suitable host cells with the expression vector or vectors, culturing the 20 transfected host cells, and recovering the CDR-grafted antibody.

The present invention also provides a method of attenuation of coagulation comprising administering a CDR-grafted antibody capable of inhibiting human tissue factor to a patient in need of such attenuation.

The present invention further provides a method of treatment or prevention of thrombotic disease comprising administering a CDR-grafted antibody capable of inhibiting human tissue factor to a patient in need 30 of such treatment or prevention. In a preferred

-7-

embodiment, the thrombotic disease is intravascular l coagulation, arterial restenosis or arteriosclerosis.

Another embodiment of the present invention is directed to a pharmaceutical composition comprising CDR-grafted antibodies capable of inhibiting human tissue factor and further comprising a pharmaceutically acceptable carrier.

# BRIEF DESCRIPTION OF THE DRAWINGS

10 Fig. 1 provides the nucleotide and deduced amino acid sequences of the heavy chain of murine monoclonal antibody TF8-5G9.

Fig. 2 provides the nucleotide and deduced amino acid sequences of the light chain of murine monoclonal antibody TF8-5G9.

Fig. 3 is a graph depicting the ability of CDR-grafted antibody TF8HCDR1 x TF8LCDR1 to bind to human tissue factor and to compete with murine monoclonal antibody TF85G9 for binding to tissue factor.

20 Solid symbols indicate direct binding of TF8HCDR1 x TF8LCDR1 and the positive control chimeric TF85G9 to tissue factor. Open symbols indicate competition binding of TF8HCDR1 x TF8LCDR1 or chimeric TF85G9 with murine monoclonal antibody TF85G9.

Fig. 4 presents the DNA sequence of expression vector pEe6TF8HCDR20 and the amino acid sequence of the coding regions of the CDR-grafted heavy chain TF8HCDR20.

Fig. 5 presents the DNA sequence of expression vector pEel2TF8LCDR3 and the amino acid sequence of the 30 coding regions of the CDR-grafted light chain TF8LCDR3.

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-8-

Fig. 6 is a graph depicting the ability of 1 CDR-grafted antibody TF8HCDR20 x TF8LCDR3 to bind to human tissue factor.

Fig. 7 is a graph depicting the ability of CDR-grafted antibody TF8HCDR20 x TF8LCDR3 to compete 5 with murine monoclonal antibody TF85G9 for binding to tissue factor.

Fig. 8 is a graph depicting the ability of CDR-grafted antibody TF8HCDR20 x TF8LCDR3 to inhibit factor X activation.

Fig. 9 provides expression vector pEe6TF8HCDR20 resulting from the subcloning of CDR-grafted heavy chain TF8HCDR20 into myeloma expression vector pEehCMV-BqlI. The following abbreviations are used: VH is the CDR-grafted heavy chain variable region; Cγ4 is the human IgG4 constant region; pA is the polyadenylation signal; ampR is the β-lactamase gene; and hCMV is human cytomegalovirus.

Fig. 10 provides expression vector
pEe12TF8LCDR3 resulting from the subcloning of CDR20 grafted light chain TF8LCDR3 into myeloma expression
vector pEe12. The following abbreviations are used: VL
is the CDR-grafted light chain variable region; CK is
the human kappa constant region; SVE is the SV40 early
promoter; GS is glutamine synthetase cDNA. Other
25 abbreviations are as noted in Fig. 9.

# DETAILED DESCRIPTION OF THE INVENTION

The present invention provides CDR-grafted
30 antibodies capable of inhibiting human tissue factor
wherein the CDRs are derived from a non-human monoclonal

antibody against tissue factor and the FR and C regions l are derived from one or more human antibodies. The present invention further provides methods of making and using the subject CDR-grafted antibodies.

In accordance with the present invention, the 5 CDR-grafted antibody is an antibody in which the CDRs are derived from a non-human antibody capable of binding to and inhibiting the function of human tissue factor, and the FR and C regions of the antibody are derived from one or more human antibodies. The CDRs derived 10 from the non-human antibody preferably have from about 90% to about 100% identity with the CDRs of the nonhuman antibody, although any and all modifications, including substitutions, insertions and deletions, are contemplated so long as the CDR-grafted antibody 15 maintains the ability to bind to and inhibit tissue factor. The regions of the CDR-grafted antibodies that are derived from human antibodies need not have 100% identity with the human antibodies. In a preferred embodiment, as many of the human amino acid residues as 20 possible are retained in order than immunogenicity is negligible, but the human residues, in particular residues of the FR region, are substituted as required and as taught hereinbelow in accordance with the present invention. Such modifications as disclosed herein are 25 necessary to support the antigen binding site formed by the CDRs while simultaneously maximizing the humanization of the antibody.

Non-human monoclonal antibodies against human tissue factor from which the CDRs can be derived are 30 known in the art (Ruf et al., 1991; Morrisey et al., 1988, Thrombosis Research 52:247) or can be produced by

-10-

well-known methods of monoclonal antibody production
l (see, e.g. Harlow et al., eds., 1988, Antibodies, A
 Laboratory Manual, Cold Spring Harbor Laboratories, Cold
 Spring Harbor, New York). Purified human tissue factor
 against which monoclonal antibodies can be raised is
5 similarly well-known (Morrisey et al., 1987, Cell
 50:129) and available to the skilled artisan. Murine

50:129) and available to the skilled artisan. Murine monoclonal antibodies, and in particular murine monoclonal antibody TF8-5G9 disclosed by Ruf et al. and Morrisey et al., 1988, Thrombosis Research 52:247, and U.S. Patent No. 5,223,427 are particularly preferred.

The ordinarily skilled artisan can determine the sequences of the CDRs by reference to published scientific literature or sequence databanks, or by cloning and sequencing the heavy and light chains of the antibodies by conventional methodology. In accordance with the present invention, the cDNA and amino acid sequences of the heavy chain (SEQ ID NOS:1 and 2, respectively) and light chain (SEQ ID NOS:3 and 4, respectively) of murine monoclonal antibody TF8-5G9 are provided. The cDNA and deduced amino acid sequence of the murine TF8-5G9 heavy chain is provided at Figure 1. The cDNA and deduced amino acid sequence of the murine TF8-5G9 light chain is provided at Figure 2.

Each of the heavy and light chain variable
25 regions contain three CDRs that combine to form the
antigen binding site. The three CDRs are surrounded by
four FR regions that primarily function to support the
CDRs. The sequences of the CDRs within the sequences of
the variable regions of the heavy and light chains can
30 be identified by computer-assisted alignment according
to Kabat et al. (1987) in Sequences of Proteins of

Immunological Interest, 4th ed., United States
1 Department of Health and Human Services, US Government
Printing Office, Washington, D.C., or by molecular
modeling of the variable regions, for example utilizing
the ENCAD program as described by Levitt (1983) J. Mol.
5 Biol. 168:595.

In a preferred embodiment the CDRs are derived from murine monoclonal antibody TF8-5G9. The preferred heavy chain CDRs have the following sequences:

10	CDR1	DDYMH	(SEQ ID NO:5)
	CDR2	LIDPENGNTIYDPKFQG	(SEQ ID NO:6)
	CDR3	DNSYYFDY	(SEQ ID NO:7)

The preferred light chain CDRs have the following sequences:

CDR1	KASQDIRKYLN	(SEQ	ID	NO:8)
CDR2	YATSLAD	(SEQ	ID	NO:9)
CDR3	LOHGESPYT	(SEO	ID	NO:10)

20

The sequences of the CDRs of the murine or other non-human antibody, and in particular the sequences of the CDRs of TF8-5G9, may be modified by insertions, substitutions and deletions to the extent that the CDR-grafted antibody maintains the ability to bind to and inhibit human tissue factor. The ordinarily skilled artisan can ascertain the maintenance of this activity by performing the functional assays described hereinbelow. The CDRs can have, for example, from about 50% to about 100% homology to the CDRs of SEQ ID NOS:5-10. In a preferred embodiment the CDRs have from about

-12-

80% to about 100% homology to the CDRs of SEQ ID NOS:5-1 10. In a more preferred embodiment the CDRs have from about 90% to about 100% homology to the CDRs of SEQ ID NOS:5-10. In a most preferred embodiment the CDRs have from about 100% homology to the CDRs of SEQ ID NOS:5-10.

The FR and C regions of the CDR-grafted antibodies of the present invention are derived from one or more human antibodies. Human antibodies of the same class and type as the antibody from which the CDRs are derived are preferred. The FR of the variable region of 10 the heavy chain is preferably derived from the human antibody KOL (Schmidt et al., 1983, Hoppe-Seyler's Z. Physiol. Chem. 364:713) The FR of the variable region of the light chain is preferably derived from the human antibody REI (Epp et al., 1974, Eur. J. Biochem.

15 45:513). In accordance with the present invention, it has been discovered that certain residues of the human FR are preferably replaced by the corresponding residue of the non-human antibody from which the CDRs are derived. For example, certain FR residues of TF8-5G9 20 are preferably retained to achieve optimal binding to antigen.

For convenience, the numbering scheme of Kabat et al. has been adopted herein. Residues are designated by lower case numbers or hyphens as necessary to conform 25 the present sequences to the standard Kabat numbered sequence.

In accordance with the present invention, residues that are retained in the FR region, i.e residues that are not replaced by human FR residues, are 30 determined according to the following guidelines. Residues that are idiosyncratic to the parent antibody,

e.g. TF8-5G9, relative to a human consensus sequence of 1 Kabat et al, are retained. Residues of the parent antibody that are in agreement with the consensus sequence are retained if the corresponding residue of the human antibody, e.g. KOL or REI, is idiosyncratic.

5 Residues that are part of the antibody loop canonical structures defined by Chothia et al. (1989) Nature 342:877, such as residue 71 of the heavy and light chains, are retained. FR residues predicted to form loops, such as residues 28-30 of the heavy chain, are

10 retained. FR residues predicted to influence the conformation of the CDRs such as residues 48 and 49 preceding CDR2 of the heavy chain, are retained.

Residues that have been demonstrated to be critical in the humanization of other antibodies may also be

15 retained. The foregoing guidelines are followed to the extent necessary to support the antigen binding site formed by the CDRs while simultaneously maximizing the humanization of the antibody.

The amino acid sequence of a representative 20 CDR-grafted heavy chain variable region derived from murine monoclonal antibody TF8-5G9 and human antibody KOL is shown below. The CDR-grafted heavy chain is designated TF8HCDR1; murine residues were retained in the FR at residues 6, 17, 23, 24, 28, 29, 30, 48, 49,

 10
 20
 30
 35ab
 50

 QVQLVQSGGG
 VVQPGRLLRL
 SCKASGFNIK
 DYYMH--WVR
 QAPGKGLEWIG

 52abc
 60
 70
 80
 82abc
 90

 LIDP--ENGNTIYD
 PKFQGRFSIS
 ADTSK--NTAFL
 QMDSLRPEDTAVY

 100
 110

25 68, 71, 73, 78, 88 and 91. CDRs are underlined.

30 YCARDNSYYF DYWGQGTPVT VSS (SEQ ID NO:11)

The amino acid sequence of a representative 1 CDR-grafted light chain variable region derived from murine monoclonal antibody TF8-5G9 and human antibody REI is shown below. The CDR-grafted light chain is designated TF8LCDR1; murine residues were retained in 5 the FR at residues 39, 41, 46 and 105. CDRs are underlined.

10 20 30 40 50

DIQMTQSPSS LSASVGDRVT ITCKASQDIR KYLNWYQQK WKAPKTLIYY

10 60 70 80 90 100

ATSLADGVPS RFSGSGSGTD YTFTISSLQP EDIATYYCLQ HGESPYTFGQ

GTKLEITR (SEQ ID NO:12)

15 regions TF8HCDR1 and TF8LCDR1 has been demonstrated in accordance with the present invention to be as effective as murine monoclonal antibody TF8-5G9 in binding to human tissue factor. It has been further discovered in accordance with the present invention, by examination of the molecular structure of murine monoclonal antibody TF8-5G9, and by design, construction, and analysis of CDR-grafted antibodies, that the FR regions can be further humanized without the loss of antigen binding activity. In particular, the FR region may retain the human FR residue at residues 6, 17, 68, 73 and 78 of the heavy chain, and residues 39, 41, 16 and 105 of the light chain, with maintenance of antigen binding activity.

In a most preferred embodiment, the heavy

30 chain variable region contains a FR derived from human antibody KOL in which murine monoclonal antibody TF8-5G9

PCT/US96/09287 WO 96/40921

-15-

residues are retained at amino acids 23, 24, 28, 29, 30, 1 48, 49, 71, 88 and 91. The preferred heavy chain variable region is designated TF8HCDR20 and has the following sequence.

10 20 5 30 35ab 50 QVQLVESGGG VVQPGRSLRL SCKASGFNIK DYYMH--WVR QAPGKGLEWIGL

52abc 60 80 82abc 90 100 IDP--ENGNTIYD PKFQGRFTIS ADNSKNTLFL QMDSLRPEDTAVY YCARDNSYYF

110 10 DYWGQGTPVT VSS (SEQ ID NO:13)

> In a most preferred embodiment, the light chain variable region contains a FR derived from human antibody REI in which murine monoclonal antibody TF8-5G9 residues are retained at amino acids 39 and 105. The preferred light chain variable region is designated TF8LCDR20 and has the following sequence.

10 - 20 30 50 DIQMTQSPSS LSASVGDRVT ITCKASQDIR KYLNWYQQKP GKAPKLLIYY 20. 70 80 ATSLADGVPS RFSGSGSGTD YTFTISSLQP EDIATYYCLQ HGESPYTFGQ GTKLEITR (SEQ ID NO:14)

It is within the ken of the ordinarily skilled. artisan to make minor modifications of the foregoing sequences, including amino acid substitutions, deletions. and insertions. Any such modifications are within the scope of the present invention so long as the resulting CDR-grafted antibody maintains the ability to bind to and inhibit human tissue factor. The ordinarily skilled 30 artisan can assess the activity of the CDR-grafted

antibody with reference to the functional assays l described hereinbelow.

The human constant region of the CDR-grafted antibodies of the present invention is selected to minimize effector function. The intended use of the 5 CDR-grafted antibodies of the present invention is to block the coagulation cascade by inhibition of tissue factor, and thus antibody effector functions such as fixation of complement are not desirable. Antibodies with minimal effector functions include IgG2, IgG4, IgA, IgD and IgE. In a preferred embodiment of the present invention, the heavy chain constant region is the human IgG4 constant region, and the light chain constant region is the human IgG4 kappa constant region.

In that effector functions may not be

desirable for therapeutic uses, the present invention
further contemplates active fragments of the CDR-grafted
antibodies, and in particular Fab fragments and F(ab')<sub>2</sub>
fragments. Active fragments are those fragments capable
of inhibiting human tissue factor. Fab fragments and

F(ab')<sub>2</sub> fragments may be obtained by conventional means,
for example by cleavage of the CDR-grafted antibodies of
the invention with an appropriate proteolytic enzyme
such as papain or pepsin, or by recombinant production.
The active fragments maintain the antigen binding sites
of the CDR-grafted antibodies and thus are similarly
useful therapeutically.

The ability of the CDR-grafted antibodies designed and constructed as taught in accordance with the present invention to bind and inhibit human tissue 30 factor can be assessed by functional assays. For example, in a rapid and convenient assay, expression

vectors containing nucleic acids encoding the CDR
1 grafted heavy and light chains can be co-transfected into suitable host cells and transiently expressed. The resulting antibodies can be assessed by standard assays for ability to bind human tissue factor, and for ability to compete for binding to tissue factor with the non-human antibody from which the CDRs are derived.

For example, transient expression of nucleic acids encoding the CDR-grafted heavy and light chains in COS cells provides a rapid and convenient system to test antibody gene expression and function. Nucleic acids encoding the CDR-grafted heavy and light chains, respectively, are cloned into a mammalian cell expression vector, for example pSG5, described by Green et al. (1988) Nucleic Acids Res. 16:369 and commercially available from Stratagene Cloning Systems, La Jolla, CA. The pSG5 expression vector provides unique restriction sites for the insertion of the heavy and light chain genes, and in vivo expression is under the control of the SV40 early promoter. Transcriptional termination is signaled by the SV40 polyadenylation signal sequence.

The pSG5-based expression vectors containing nucleic acids encoding the heavy and light chains are cotransfected into COS cells and cultured under conditions suitable for transient expression. Cell culture media is then harvested and examined for antibody expression, for example by an enzyme linked immunosorbent assay (ELISA), to determine that suitable levels of antibody have been produced. An ELISA may then be used to assess the ability of the CDR-grafted antibody to bind to human tissue factor. Human tissue factor is immobilized on a microtiter plate and the COS

cell supernatant containing the CDR-grafted antibody is

1 added followed by an incubation at room temperature for
about one hour. The plates are then washed with a
suitable detergent-containing buffer such as phosphate
buffered saline (PBS)/Tween, followed by the addition of

5 the components of a suitable detection system. For
example, horseradish peroxidase conjugated goat antihuman kappa chain polyclonal antibody is added, followed
by washing, followed by addition of substrate for
horseradish peroxidase, and detection. The CDR-grafted

10 antibodies within the scope of the present invention are
those which are capable of binding to human tissue
factor to a degree comparable to the non-human antibody
from which the CDRs are derived as determined by the
foregoing assay.

15 The ability of the CDR-grafted antibodies to inhibit the activity of human tissue factor in vivo can be conveniently assessed by the following in vitro assay that mimics in vivo coagulation events. In response to vascular injury in vivo, tissue factor binds to factor 20 VII and facilitates the conversion of factor VII to a serine protease (factor VIIa). The factor VIIa-tissue factor complex converts factor X to a serine protease (factor Xa). Factor Xa forms a complex with factor Va (from the intrinsic coagulation pathway), resulting in 25 the conversion of prothrombin to thrombin, which in turn results in the conversion of fibrinogen to fibrin. convenient in vitro functional assay, tissue factor is incubated in the presence of factor VIIa and the CDRgrafted anti-tissue factor antibody produced in the transient expression system described above. Factor X is added and the reaction mixture is incubated, followed

-19-

by an assay for factor Xa activity utilizing a

1 chromogenic substrate for factor Xa (Spectrozyme FXa,
American Diagnostica, Inc., Greenwich, CT). The ability
of the CDR-grafted antibody to inhibit factor X
activation thus provides a measure of the ability of the
5 CDR-grafted antibody to inhibit the activity of human
tissue factor.

The CDR-grafted antibodies within the scope of the present invention are those which are capable of inhibiting human tissue factor to a degree comparable to 10 the non-human antibody from which the CDRs are derived as determined by the foregoing assay. In one embodiment, the CDR-grafted antibody has at least 50% of the inhibitory activity of TF8-5G9 for human tissue factor. In a preferred embodiment, the CDR-grafted 15 antibody has at least 70% of the inhibitory activity of TF8-5G9 for human tissue factor. In a more preferred embodiment, the CDR-grafted antibody has at least 80% of the inhibitory activity of TF8-5G9 for human tissue factor. In a most preferred embodiment, the CDR-grafted 20 antibody has at least 90% of the inhibitory activity of TF8-5G9 for human tissue factor.

In another embodiment, the present invention provides a method of producing a CDR-grafted antibody capable of inhibiting human tissue factor. The method comprises constructing an expression vector containing a nucleic acid encoding the CDR-grafted antibody heavy chain and an expression vector containing a nucleic acid encoding the CDR-grafted antibody light chain, transfecting suitable host cells with the expression vectors, culturing the transfected host cells under conditions suitable for the expression of the heavy and

light chains, and recovering the CDR-grafted antibody.

1 Alternately, one expression vector containing nucleic acids encoding the heavy and light chains may be utilized.

Standard molecular biological techniques, for 5 example as disclosed by Sambrook et al. (1989), Molecular Cloning: A Laboratory Manual Cold Spring Harbor Press, Cold Spring Harbor, NY may be used to obtain nucleic acids encoding the heavy and light chains of the CDR-grafted antibodies of the present invention. 10 A nucleic acid encoding the CDR-grafted variable domain may be constructed by isolating cDNA encoding the antibody to be humanized, e.g. murine monoclonal antibody TF8-5G9, by conventional cloning methodology from the hybridoma producing the antibody, or by 15 polymerase chain reaction (PCR) amplification of the variable region genes, as described for example by Winter et al., followed by site-directed mutagenesis to substitute nucleotides encoding the desired human residues into the FR regions. Alternately, the cDNA 20 encoding the human antibody can be isolated, followed by site-directed mutagenesis to substitute nucleotides encoding the desired murine residues into the CDRs.

Nucleic acids encoding the CDR-grafted variable domain may also be synthesized by assembling 25 synthetic oligonucleotides, for example utilizing DNA polymerase and DNA ligase. The resulting synthetic variable regions may then be amplified by PCR. Nucleic acids encoding CDR-grafted variable domains may also be constructed by PCR strand overlap methods that are known in the art and reviewed by Owens et al.

Accordingly, having determined the desired

1 amino acid sequences of the CDR-grafted variable domains in accordance with the present invention, the ordinarily skilled artisan can obtain nucleic acids encoding the variable domains. Further, the skilled artisan is aware

5 that due to the degeneracy of the genetic code, various nucleic acid sequences can be constructed that encode the CDR-grafted variable domains. All such nucleic acid sequence are contemplated by the present invention.

The nucleic acids encoding the CDR-grafted

10 variable domains are linked to appropriate nucleic acids encoding the human antibody heavy or light chain constant region. Nucleic acid sequences encoding human heavy and light chain constant regions are known in the art. It is within the ken of the ordinarily skilled

15 artisan to include sequences that facilitate transcription, translation and secretion, for example start codons, leader sequences, the Kozak consensus sequence (Kozak, 1987, J. Mol. Biol. 196:947) and the like, as well as restriction endonuclease sites to

20 facilitate cloning into expression vectors.

The present invention thus further provides nucleic acids encoding the heavy and light chains of CDR-grafted antibodies capable of inhibiting human tissue factor wherein the CDRs are derived from a murine monoclonal antibody against tissue factor and the FR and C regions are derived from one or more human antibodies.

In accordance with the present invention, representative nucleic acids encoding CDR-grafted heavy and light chains were constructed. The CDR-grafted

30 heavy chain comprises a variable region containing FR regions derived from human antibody KOL and CDRs derived

- from murine monoclonal antibody TF8-5G9 and further

  comprises a constant region derived from the heavy chain of human IgG4. The CDR-grafted light chain comprises a variable region containing FR regions derived from human antibody REI and CDRs derived from murine monoclonal
- 5 antibody TF8-5G9 and further comprises a constant region derived from human IgG4 kappa chain. Nucleic acids encoding the heavy and light chains were constructed by assembling the variable regions from synthetic nucleotides, amplifying the assembled variable regions
- 10 by PCR, purifying the amplified nucleic acids, and ligating the nucleic acid encoding the variable region into a vector containing a nucleic acid encoding the appropriate human constant region.

The sequences of representative nucleic acids encoding CDR-grafted heavy and light chains are presented as nucleotides 1-2360 of SEQ ID NO:15 and nucleotides 1-759 of SEQ ID NO:20, respectively.

The nucleic acid sequence encoding a preferred heavy chain (nucleotides 1-2360 of SEQ ID NO:15) is

20 designated the TF8HCDR20 gene. The nucleic acid sequence contains the following regions: 5' EcoRI restriction site (nucleotides 1-6); Kozak sequence (nucleotides 7-15); start codon and leader sequence (nucleotides 16-72); CDR-grafted variable region

25 (nucleotides 73-423); human IgG4 CH1 domain (nucleotides 424-717); human IgG4 intron 2 (nucleotides 718-1110); human IgG4 hinge (nucleotides 1111-1146); human IgG4 intron 3 (nucleotides 1147-1267); human IgG4 CH2 domain (nucleotides 1268-1594); human IgG4 intron 4

30 (nucleotides 1595-1691); human IgG4 CH3 domain

(nucleotides 1692-2012); 3' untranslated region

-23-

(nucleotides 2013-2354); 3' <u>BamHI</u> end spliced to <u>BclI</u> site of expression vector (nucleotides 2355-2360).

The nucleic acid sequence encoding a preferred light chain gene (nucleotides 1-759 of SEQ ID NO:20) is designated the TF8LCDR3 gene. The nucleic acid sequence 5 contains the following regions: 5' EcoRI restriction site (nucleotides 1-5); Kozak sequence (nucleotides 6-8); start codon and leader sequence (nucleotides 9-68); CDR-grafted variable region (nucleotides 69-392); human kappa constant region (nucleotides 393-710); 3' Untranslated region (nucleotides 711-753); 3' BamHI end spliced to BclI site of expression vector (nucleotides 754-759).

The foregoing preferred sequences can be modified by the ordinarily skilled artisan to take into account degeneracy of the genetic code, and to make additions, deletions, and conservative and nonconservative substitutions that result in a maintenance of the function of the nucleic acid, i.e. that it encodes a heavy or light chain of a CDR-grafted antibody capable of inhibiting human tissue factor. Restriction sites and sequences that facilitate transcription and translation may be altered or substituted as necessary depending upon the vector and host system chosen for expression.

25 Suitable expression vectors and hosts for production of the CDR-grafted antibodies of the present invention are known to the ordinarily skilled artisan. The expression vectors contain regulatory sequences, such as replicons and promoters, capable of directing replication and expression of heterologous nucleic acids sequences in a particular host cell. The vectors may

-24-

also contain selection genes, enhancers, signal

1 sequences, ribosome binding sites, RNA splice sites,
polyadenylation sites, transcriptional terminator
sequences, and so on. The vectors may be constructed by
conventional methods well-known in the art, or obtained
from commercial sources. The expression vectors
preferably have convenient restriction sites at which
the nucleic acids encoding the antibody chains of the
invention are inserted. Myeloma expression vectors in
which antibody gene expression is driven by the human
cytomegalovirus promoter-enhancer or are particularly
preferred.

Expression vectors containing a nucleic acid encoding the CDR-grafted heavy chain under the control of a suitable promoter and expression vectors containing a nucleic acid encoding the CDR-grafted light chain under the control of a suitable promoter are cotransfected into a suitable host cell. In another embodiment, nucleic acids encoding both heavy and light chains are provided in a single vector for transfection of a suitable host cell.

Suitable host cells or cell lines for expression of the CDR-grafted antibodies of the present invention include bacterial cells, yeast cells, insect cells, and mammalian cells such as Chinese hamster ovary (CHO) cells, COS cells, fibroblast cells and myeloid cells. Mammalian cells are preferred. CHO, COS and myeloma cells are particularly preferred. Myeloma cells are preferred for establishing permanent CDR-grafted antibody producing cell lines. Expression of antibodies in myeloma cells, bacteria, and yeast is reviewed by

above.

Sandhu (1992) Critical Reviews in Biotechnology 12:437. 1 Expression in mammalian cells is reviewed by Owen et al. Transfection of host cells by the expression vectors containing nucleic acids encoding the CDRgrafted heavy and light chains can be accomplished by 5 methods well-known to one of ordinary skill in the art. Such methods include, for example, calcium chloride transfection, calcium phosphate transfection, lipofection and electroporation. Suitable culture methods and conditions for the production of the CDR-10 grafted antibodies are likewise well-known in the art. The CDR-grafted antibodies can be purified by conventional methods, including ammonium sulfate precipitation, affinity chromatography, gel electrophoresis, and the like. The ability of the CDR-15 grafted antibodies to bind to and inhibit human tissue factor can be assessed by the in vitro assays described

The CDR-grafted antibodies of the present invention have a variety of utilities. For example, the 20 antibodies are capable of binding to human tissue factor and thus are useful in assays for human tissue factor from body fluid samples, purification of human tissue factor, and so on.

The CDR-grafted antibodies of the present
invention are capable of inhibiting human tissue factor.
Human tissue factor is well-known to be an essential
element in the human coagulation cascade. The ability
of the antibodies of the present invention to disrupt
the coagulation cascade is demonstrated by in vitro
assays in which the antibodies prevent factor X
activation. Accordingly, the present antibodies are

-26-

useful in the attenuation of coagulation. The present invention thus provides a method of attenuation of coagulation comprising administering a therapeutically effective amount of CDR-grafted antibody capable of inhibiting human tissue factor to a patient in need of such attenuation.

Numerous thrombotic disorders are characterized by excessive or inappropriate coagulation and are effectively treated or prevented by administration of agents that interfere with the coagulation cascade. Accordingly, the present invention further provides a method of treatment or prevention of a thrombotic disorder comprising administering a therapeutically effective amount of a CDR-grafted antibody capable of inhibiting human tissue factor to a patient in need of such treatment or prevention. In a preferred embodiment, the thrombotic disorder is intravascular coagulation, arterial restenosis or arteriosclerosis. The antibodies of the invention may be used in combination with other antibodies or therapeutic agents.

A therapeutically effective amount of the antibodies of the present invention can be determined by the ordinarily skilled artisan with regard to the patient's condition, the condition being treated, the 25 method of administration, and so on. A therapeutically effective amount is the dosage necessary to alleviate, eliminate, or prevent the thrombotic disorder as assessed by conventional parameters. For example, a therapeutically effective dose of a CDR-grafted antibody of the present invention may be from about 0.1 mg to about 20 mg per 70 kg of body weight. A preferred

dosage is about 1.0 mg to about 5 mg per 70 kg of body 1 weight.

A patient in need of such treatment is a patient suffering from a disorder characterized by inappropriate or excessive coagulation, or a patient at risk of such a disorder. For example, anticoagulant therapy is useful to prevent postoperative venous thrombosis, and arterial restenosis following balloon angioplasty.

The CDR-grafted antibodies of the present

10 invention are useful in the same manner as comparable therapeutic agents, and the dosage level is of the same order of magnitude as is generally employed with those comparable therapeutic agents. The present antibodies may be administered in combination with a

15 pharmaceutically acceptable carrier by methods known to one of ordinary skill in the art.

Another embodiment of the present invention is directed to a pharmaceutical composition comprising a least one CDR-grafted antibody capable of inhibiting 20 human tissue factor and further comprising a pharmaceutically acceptable carrier. As used herein, "pharmaceutically acceptable carrier" includes any and all solvents, dispersion media, coatings, antibacterial and antifungal agents, isotonic and absorption delaying 25 agents, and the like. The use of such media and agents for pharmaceutically active substances is well-known in the art. Except insofar as any conventional media or agent is incompatible with the active ingredient, its use in the therapeutic compositions is contemplated.

30 Supplementary active ingredients can also be

incorporated into the compositions.

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The antibodies can be administered by wellknown routes including oral and parenteral, e.g.,
intravenous, intramuscular, intranasal, intradermal,
subcutaneous, and the like. Parenteral administration
and particularly intravenous administration is
preferred. Depending on the route of administration,
the pharmaceutical composition may require protective
coatings.

The pharmaceutical forms suitable for injectionable use include sterile aqueous solutions or 10 dispersions and sterile powders for the extemporaneous preparation of sterile injectable solutions or dispersions. In all cases the ultimate solution form must be sterile and fluid. Typical carriers include a solvent or dispersion medium containing, for example, 15 water buffered aqueous solutions (i.e., biocompatible buffers), ethanol, polyol such as glycerol, propylene glycol, polyethylene glycol, suitable mixtures thereof, surfactants or vegetable oils. The antibodies may be incorporated into liposomes for parenteral 20 administration. Sterilization can be accomplished by an art-recognized techniques, including but not limited to, addition of antibacterial or antifungal agents, for example, paraben, chlorobutanol, phenol, sorbic acid or thimersal. Further, isotonic agents such as sugars or 25 sodium chloride may be incorporated in the subject compositions.

Production of sterile injectable solutions containing the subject antibodies is accomplished by incorporating these antibodies in the required amount in the appropriate solvent with various ingredients enumerated above, as required, followed by

sterilization, preferably filter sterilization. To

l obtain a sterile powder, the above solutions are vacuumdried or freeze-dried as necessary.

The following examples further illustrate the present invention.

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### EXAMPLE 1

Two DNA libraries were generated from oligo

5 (dT)-primed TF8-5G9 hybridoma RNA utilizing standard
molecular biology procedures as described by Sambrook et
al. The cDNA was cloned into the Librarian II plasmid
vector from Invitrogen (San Diego, CA), and the
libraries were screened for cDNA clones encoding murine

10 IgG HC and LC. A full-length cDNA clone for the heavy
chain could not be isolated, despite the construction of
two independent libraries. A random primed TF8-5G9 cDNA
library was generated to obtain the missing 5' sequence
of the heavy chain. Consequently, the heavy chain cDNA

15 was in two pieces: a 5' clone of 390 nucleotides and a
3' clone of 1392 nucleotides. The two HC clones overlap
by 292 nucleotides.

The HC and LC clones were completely sequenced by the dideoxy chain termination method of Sanger et al.

20 (1977) Proc. Natl. Acad. Sci. USA 74:5463. To verify the variable region sequence, sequence was obtained from PCR-amplified cDNA that had been synthesized from total TF8-5G9 hybridoma RNA. Total TF8-5G9 hybridoma RNA was isolated by the guanidinium thiocyanate method of

25 Chrigwin et al. (1970) Biochemistry 18:5294. cDNA was synthesized using the Perkin Elmer (Norwalk, CT) GeneAmp RNA Polymerase Chain Reaction (PCR) kit with an oligo (dT) primer. Components of the same kit were used in the PCR to amplify the LC and HC variable regions using primers based on the sequence that had been obtained for the cDNA clones. The amplified variable region

fragments were gel-purified and sequenced according to

1 the method of Tracy et al. (1991) BioTechniques 11:68 on
a Model 373A Applied Biosystems, Inc. (Foster City, CA)
automated fluorescent DNA sequencer. The sequence for
TF8-5G9 LC and HC obtained from RNA amplification and
5 the sequence obtained from the cDNA clones agreed. The
TF8-5G9 HC variable region sequence with protein
translation is shown in Figure 1 and SEQ ID NO:1, and
that for the LC is shown in Figure 2 and SEQ ID NO:3.

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### EXAMPLE 2

l Chimeric LC and HC Expression Vector Construction

In order to test the binding activity of the CDR-grafted anti-TF LC and HC individually, mouse-human 5 chimeric TF8-5G9 LC and HC were constructed. This allowed the CDR-grafted LC to be tested for TF binding ability in combination with the chimeric HC, and the CDR-grafted HC to be tested in combination with the chimeric LC.

LC variable region using as template cDNA clones in the Librarian II vector. The 5' primer was designed with an EcoRI site while the 3' primer was designed with a NarI site. PCR was used to amplify the LC variable region, generating a 433 bp fragment with a 5'EcoRI end and 3'NarI end. The fragment included the signal sequence from the TF8-5G9 LC cDNA clone but incorporated a 2 base change in the arginine codon immediately following the ATG start codon. This change retained the arginine residue but made the sequence conform to the Kozak consensus sequence in order to potentially improve translation of the LC mRNA. The PCR amplified LC

<u>NarI</u> restriction enzymes and purified by electrophoresis on a 2% Nusieve, 1% Seakem agarose gel (FMC Bio Products, Rockland, ME).

variable region fragment was digested with EcoRI and

The DNA was extracted from the gel slice and purified by the Geneclean (Bio 101, La Jolla, CA) procedure. The full-length chimeric TF8-5G9 LC gene was generated by cloning this DNA into the EcoRI and NarI sites of a pSP73 vector (Promega, Madison, WI) which

-33-

contains the human kappa constant region. The gene was isolated from the pSP73 vector by <a href="EcoRI"><u>EcoRI</u></a> digestion and subcloned into the <a href="EcoRI"><u>EcoRI</u></a> site of the pSG5 mammalian cell expression vector (Stratagene Cloning Systems, La Jolla, CA).

The chimeric TF8-5G9 HC gene was assembled in a manner similar to that of the chimeric LC. Since there was no full-length HC cDNA isolated from the Librarian II vector cDNA libraries, the HC variable region fragment that was generated by the PCR from total TF8-5G9 hybridoma cell RNA was used as the template. Primers which incorporated an <a href="EcoRI">EcoRI</a> site at the 5' end and a <a href="SacI">SacI</a> site at the 3' end were used in the PCR to generate a 430 bp fragment which contained the TF8-5G9 HC Kozak sequence, start codon, signal sequence, and variable region. This fragment was digested with the restriction enzymes <a href="EcoRI">EcoRI</a> and <a href="SacI">SacI</a>, and gel-purified using the same procedure that was used with the chimeric LC construction.

The full-length TF8-5G9 chimeric HC gene was constructed by cloning the variable region fragment into the <a href="EcoRI">EcoRI</a> and <a href="SacI">SacI</a> sites of the pSG5 expression vector containing the human <a href="IgG4">IgG4</a> constant region.

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#### EXAMPLE 3

Design and Construction of the CDR-Grafted Heavy and Light Chain Genes

The variable region domains of the CDR-grafted 5 HC and LC genes were designed with an EcoRI overhang at the 5' end followed by a Kozak sequence to improve antibody expression. The leader sequences were derived from the heavy and light chains of the murine monoclonal antibody B72.3 (Whittle et al. (1987) Protein

10 Engineering 1:499). The 3' end of the variable regions were designed to have overhangs which allowed for splicing to the appropriate human constant region DNA.

In the initially designed CDR-grafted TF8-5G9 heavy and light chains the CDRs were derived from murine TF8-5G9 sequence while the frameworks were derived primarily from human antibody sequence. The human antibody KOL (Schmidt et al.) was used for the heavy chain frameworks, while the human antibody dimer (Epp et al.) was used for the light chain frameworks.

- Several criteria were used to select murine framework residues in the design of the TF8-5G9 CDR-grafted heavy and light chain variable regions. Framework residues which, at a particular position, are idiosyncratic to TF8-5G9 were retained as murine sequence with the assumption that they contributed to
  - its unique binding characteristics. TF8-5G9 murine residues were also retained at framework positions where they were in agreement with the human consensus sequence but where the corresponding residues in KOL or REI were
- 30 idiosyncratic. Residues that are part of antibody loop canonical structures such as residue 71 (numbering

- according to Kabat et al.) of the heavy and light chains l were also retained as murine sequence. Framework residues that form loops such as residues 26-30 of the HC were kept as TF8-5G9 murine sequence at positions were the murine sequence differed from the human.
- 5 Residues known to directly influence the conformation of CDRs, such as 48 and 49 immediately preceding CDR2 of the HC, were also retained as murine sequence.

The amino acid sequence of the variable region for the initially designed CDR-grafted TF8-5G9 HC,

10 TF8HCDR1, is shown in SEQ ID NO:11. Murine residues were retained at framework positions 6, 17, 23, 24, 28, 29, 30, 48, 49, 68, 71, 73, 78 88 and 91. The CDR-grafted HC variable region was attached to a human IgG4 constant region.

The amino acid sequence of the variable region for the initially designed CDR-grafted TF8-5G9 LC, TF8LCDR1, is shown in SEQ ID NO:12. Murine residues were retained at framework positions 39, 41, 46 and 105. The CDR-grafted LC variable region was attached to a 20 human kappa constant region.

The variable region for the CDR-grafted HC and LC described above were each assembled from 13 synthetic oligonucleotides which were synthesized by Research Genetics, Inc., Huntsville, AL. These oligonucleotides 25 ranged in length from 42 to 80 bases, and encoded both variable region strands. When the 6 complementary oligonucleotide pairs were annealed, the overhangs generated were 17 to 24 bases in length. These oligonucleotide pairs were combined, annealed at their complementary overhangs, and ligated to give the final full length double-stranded variable regions.

WO 96/40921 PCT/US96/09287

-36-

The HC variable region oligonucleotides were

1 assembled into a 452 bp fragment which contains a 5'

EcoRI site and a 3' SacI site. The polymerase chain reaction was used to amplify this fragment. The resulting amplified DNA was purified on a 2% Nusieve, 1%

- 5 Seakem agarose gel (FMC). The appropriate size band of DNA was excised and the DNA was recovered by the Geneclean (Bio 101) procedure. The fragment was then digested with <a href="EcoRI"><u>EcoRI</u></a> and <a href="SacI">SacI</a>, and purified again by the Geneclean method. This HC variable region fragment with
- 10 EcoRI and SacI ends was cloned into the EcoRI and SacI sites of the pSport-1 vector (GIBCO-BRL Life Technologies, Gaithersburg, MD). DNA from several clones was isolated and sequenced to verify proper variable region assembly. All clones had unexpected
- 15 base changes. One clone with the fewest base changes (two mismatches at bases, 133 and 140) was selected to be corrected by site-directed mutagenesis according to Kunkel (1985) <a href="Proc. Natl. Acad. Sci. USA 82:488">Proc. Natl. Acad. Sci. USA 82:488</a>. Briefly, CJ236 (ung-, dut-) competent cells (Invitrogen
- 20 Corporation, San Diego, CA) were transformed with the pSport vector containing the CDR-grafted HC variable region with the two base mismatch. Single-stranded, uridine-incorporated DNA templates were purified from phage following M13 helper phage (Stratagene Cloning
- 25 Systems) infection of the transformed cells.

  Mutagenesis oligos containing the desired base changes
  were synthesized on an Applied Biosystems Model 380B DNA
  synthesizer. The mutagenesis oligos were annealed to
  the template DNA, and T7 DNA Polymerase and T4 DNA
- 30 Ligase (MutaGene InVitro Mutagenesis Kit, Bo-Rad Laboratories, Richmond, CA) were used to incorporate the

PCT/US96/09287 WO 96/40921 -37-

oligo into a newly synthesized DNA strand. l competent cells (GIBCO-BRL Life Technologies) were transformed with the double-stranded DNA. The original uridine-incorporated strand is destroyed while the newly synthesized strand containing the mutagenesis oligo is 5 replicated. Phagemid DNA was prepared from the

resulting mutagenesis clones and the variable regions were sequence to identify the clones which had incorporated the desired changes. The corrected HC EcoRI/SacI variable region fragment was excised from the 10 pSport vector, purified and ligated into the EcoRI/SacI sites of a pSG5 vector containing the human IgG4

constant region. This resulted in the generation of a full-length humanized TF8-5G9 HC gene, TF8HCDR1, in the pSG5 COS cell expression vector. The vector was

15 designated pSG5TF8HCDR1.

The CDR-grafted TF8-5G9 LC variable region was also amplified by the PCR from the assembled synthetic oligonucleotides into a 433 bp fragment which contained a 5' EcoRI site and a 3' NarI site. This fragment was 20 purified as described above for the HC, digested with EcoRI and NarI and purified by the Geneclean procedure. This fragment was cloned into the EcoRI and NarI sites of a pSG5 vector which contains the human kappa constant region. This resulted in the generation of a full-25 length humanized TF8-5G9 LC gene, TF8LCDR1, in the pSG5 COS cell expression vector. Seven clones were sequenced, and one was found to have the desired CDRgrafted LC sequence. The vector was designated

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pSQ5TF8LCDR1.

WO 96/40921 PCT/US96/09287

-38-

## EXAMPLE 4

Expression of the CDR-Grafted
Heavy and Light Chain Genes in COS Cells

The transient expression of antibody genes in

5 COS-1 cells provides a rapid and convenient system to
test antibody gene expression and function. COS-1 cells
were obtained from the American Type Culture Collection
(CRL 1650) and cultured in Dulbecco's Modified Eagle
Medium (DMEM, from GIBCO BRL Life Technologies) with 10%

10 fetal calf serum. The pSG5TF8HCDR1 expression factor
was cotransfected into COS cells with the pSG5 chimeric
LC expression vector using the DEAE-Dextran method
followed by DMSO shock as described by Lopata et al.
(1984) Nucleic Acids Res. 14:5707. After 4 days of

15 culture, media was harvested from the wells and examined
for antibody expression levels.

Antibody levels were determined by an ELISA-based assembly assay. Plates were coated with a goat anti-human Fc specific antibody. Various dilutions of the COS cell supernatant containing secreted antibody were added, incubated for one hour, and washed. A horseradish peroxidase-linked goat anti-human kappa chain antibody was added, incubated for one hour at room temperature, and washed. Substrate for the horseradish peroxidase was added for detection. Antibody levels in the COS cell media were found to be nearly undetectable for the TF8HCDR1 x chimeric LC. Upon closer examination of the TF8HCDR1 variable region sequence, it was found that an unexpected base change, which had occurred during the site-directed mutagenesis process described in Example 3, introduced a stop codon into framework 4

- of the TF8HCDR1 gene. This substitution was corrected

  by site-directed mutagenesis as described above.

  Thorough sequencing of the variable region confirmed that the correction was made with no additional changes introduced. Upon transfection of this corrected
- 5 TF8HCDR1 gene with the chimeric LC, reasonable expression levels were obtained.

COS cells which had been co-transfected with the CDR-grafted LC expression vector, pSGTF8LCDR1, and either the chimeric HC or TF8HCDR1, produced antibody at reasonable levels. Antibody levels in COS cell supernatants ranged from 0.5  $\mu$ g to 10.0  $\mu$ g per ml.

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### EXAMPLE 5

Binding of the CDR-Grafted TF8-5G9 to Tissue Factor

An ELISA was used to determine the ability of the CDR-grafted TF8-5G9 antibody, TF8HCDR1 x TF8LCDR1,

5 to bind to tissue factor. Tissue factor was immobilized on a microtiter plate. The test COS cell supernatant, containing the CDR-grafted antibody, was added to the well, incubated for one hour at room temperature. Following three washes with PBS/Tween, a goat anti-human lo kappa chain polyclonal antibody conjugated to horseradish peroxidase was added, incubated for one hour at room temperature and washed. Substrate for the horseradish peroxidase was added for detection. The positive control was the TF8-5G9 chimeric antibody. The CDR-grafted TF8-5G9 antibody was able to bind to tissue factor to a degree comparable to the chimeric TF8-5G9 antibody (Figure 3, solid symbols).

The ability of the humanized antibody to compete with murine TF8-5G9 for binding to tissue factor 20 was also examined. Varying amounts of COS cell supernatant containing the test CDR-grafted antibody and a fixed amount of murine TF8-5G9 were added simultaneously to wells coated with tissue factor. Binding was allowed to occur for one hour at room 25 temperature. The wells were washed three times with PBS/Tween. A goat anti-human kappa chain antibody conjugated to horseradish peroxidase was added, incubated for one hour at room temperature and washed. Substrate for the horseradish peroxidase was added for detection. The positive antibody competed as well as

the chimeric antibody with murine TF8-5G9 for binding to 1 TF.

These data indicate that the initially designed CDR-grafted antibody, TF8HCDR1 x TF8LCDR1, was approximately as active as the chimeric TF8-5G9 in 5 binding to TF and competing with the murine antibody for binding to TF.

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## EXAMPLE 6

Construction and Characterization
of Additional CDR-Grafted Heavy Chains

Upon examination of the molecular structure of 5 murine TF8-5G9, framework residues at positions 27, 68, 73 and 78 were found to lie on the antibody surface and had no discernible contact with the CDRs. framework residues were of murine sequence in TF8HCDR1 but were changed to the human KOL sequence in various 10 combinations to generate a series of CDR-grafted heavy. chains with framework residue variations. The changes were made by the process of site-directed mutagenesis as described in Example 3. Each CDR-grafted heavy chain version was expressed in COS cells in combination with 15 the CDR-grafted LC, TF8LCDR1, and tested for its ability to bind TF and compete with murine TF8-5G9 for binding. Every version of the CDR-grafted heavy chain in combination with TF8LCDR1 was shown to bind TF with an affinity comparable to chimeric TF8-5G9. Every CDR-20 grafted HC in combination with TF8LCDR1 was able to compete with murine TF8-5G9 for binding to TF to a degree comparable to the chimeric antibody.

Changes in sequence from murine to human for HC framework positions 6, 7, 68, 73 and 78 did not 25 adversely affect the antigen binding ability of the antibody. The CDR-grafted HC version which had human sequence at all of these positions, and thus was the most humanized HC, was TF8HCDR20.

The complete sequence of the TF8HCDR20 gene
30 was determined. The DNA sequence is shown as a 2360 bp
EcoRI/BamHI insert with protein translation in the

pEe6TF8HCDR20 expression vector in Figure 4 and SEQ ID 1 NO:15.

The essential regions of the gene are as follows:

	Nucleotide #	Region
5	1-6	5' EcoRI restriction site
	7-15	Kozak sequence
	16-72	Start codon and leader sequence
	73-423	CDR-grafted variable region
	424-717	Human IgG4 CH1 domain
10	718-1110	Human IgG4 intron 2
	1111-1146	Human IgG4 hinge
,	1147-1267	Human IgG4 intron 3
	1268-1594	Human IgG4 CH2 domain
	1595-1691	Human IgG4 intron 4
15	1692-2012	Human IgG4 CH3 domain
	2013-2354	3' untranslated region
	2355-2360	3' BamHI end spliced to BclI site of the expression vector

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WO 96/40921 PCT/US96/09287

-44-

#### EXAMPLE 7

C nstruction and Characterization of Additional CDR-Grafted Light Chains

The initially designed CDR-grafted LC, 5 TF8LCDR1, contained four framework residues from the murine TF8-5G9 sequence. At two of these positions, 39 and 105, the human REI framework sequence is unique to REI; however, the murine TF8-5G9 LC sequence is in agreement with the human consensus sequence. 10 two murine framework residues, trp41 and thr46, are unique to TF8-5G9. Several versions of the CDR-grafted LC were generated in which the sequence at these four positions were changed from the murine to the human REI in various combinations. These changes were made by 15 site-directed mutagenesis. Each version of the CDRgrafted LC was expressed in COS cells in combination with the CDR-grafted HC, TF8HCDR20, and tested for ability to bind tissue factor and compete with murine TF8-5G9 for binding. Every version of the CDR-grafted 20 LC, in combination with TF8HCDR20, was shown to bind TF with an affinity comparable to TF8-5G9. Also every CDRgrafted LC version, in combination with TF8HCDR20, was able to compete with murine TF8-5G9 for binding to TF in a manner comparable to the chimeric TF8-5G9 control.

Changes in sequence from murine to human for LC framework positions 39, 41, 46 and 105 did not adversely effect the ability of the antibody to recognize antigen. The CDR-grafted LC of choice was TF8LCDR3, where murine TF8-5G9 sequence was used at positions 39 and 105 because these are in agreement with

the human consensus sequence. The preferred CDR-grafted 1 TF8-5G9 antibody is TF8HCDR20  $\times$  TF8LCDR3.

The complete sequence of the TF8LCDR3 gene was determined and is shown as a 759 bp <a href="EcoRI-BamHI">EcoRI-BamHI</a> insert with protein translation in the pEel2TF8LCDR3 expression vector in Figure 5 and SEQ ID NO:17. The essential regions of the gene are as follows:

	Nucleotide #	Region
	1-5	5' EcoRI restriction site
	6-8	Kozak sequence
10	9-68	Start codon and leader sequence
	69-392	CDR-grafted variable region
	393-710	Human kappa constant region
	711-753	3' untranslated region
	754-759	3'BamHI end spliced to BclI
15		site of the expression vector

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## EXAMPLE 8

CDR-Grafted TF8-5G9 Antibody TF8HCDR20 x TF8LCDR3
Inhibits Human Tissue Factor

The binding of the CDR-grafted TF8-5G9

5 antibody, TF8HCDR20 x TF8LCDR3, to TF was assessed as described in Example 5 and was found to be comparable to that of the chimeric TF8-5G9 as illustrated in Figure 6. The ability of the CDR-grafted TF8-5G9 to compete with the murine antibody for binding to TF is comparable to that of the chimeric TF8-5G9 as shown in Figure 7.

An <u>in vitro</u> assay was used to measure the level of inhibition of factor X activation by the CDR-grafted TF8-5G9 antibody. In this assay, TF forms an active proteolytic complex with factor VII. This

15 complex then converts factor X to factor Xa by proteolysis. The activated Xa enzymatically cleaves a substrate, Spectrozyme FXa, which releases a chromogen. The level of chromogen, as detected by optical density, is an indication of factor X activation due to TF-factor 20 VIIa activity.

The following reaction mixtures were prepared in 12 x 75 mm borosilicate glass tubes.

25  $\mu$ l TBS (50 mM Tris, pH 7.4, 150 mM NaCl) 15  $\mu$ l 20 mM CaCl<sub>2</sub>/1% bovine serum albumin

25 (BSA)

20  $\mu$ l human placental tissue factor solution (prepared by reconstituting one vial of Thromborel S, Curtin Matheson Scientific #269-338 with 4.0 ml dH<sub>2</sub>O and diluting 1:10 in TBS)

30  $\mu$ l Factor VII (Enzyme Research Labs #HFVII 1 1007 at 237.66 ng/ml in TBS) 30  $\mu$ l TBS or TF8-5G9 or TF8MCDR20 x TF8LCDR3 at 1.18  $\mu$ g/ml or as indicated in Fig. 8 The reaction mixtures were incubated at 37°C

- 5 for ten minutes before the addition of Factor X. (In some cases the reaction mixture was preincubated for five minutes before addition of Factor VII or antibody, followed by a ten minute incubation before addition of Factor X.) Thirty  $\mu$ l of Factor X solution (Enzyme
- Research Labs, DHFX 330, 247.38  $\mu$ g/ml TBS) was added and the mixture was incubated at 37°C for three minutes. Factor X activation was terminated by pipetting 40  $\mu$ g of reaction mixture into 160  $\mu$ l of stop buffer (50 mM Tris, pH 7.4, 100 mM EDTA, 150 mM NaCl) in 96 well microtiter
- 15 plates. Each tube of reaction mixture was pipetted into three microtiter wells. Fifty μl of Spectrozyme FXa substrate (American Diagnostica #222, 1μM/ml TBS) was added to each well. OD<sub>405</sub> was read on a Molecular Devices kinetic plate reader with readings taken every twenty seconds for ten minutes. Factor X activity was
- 20 twenty seconds for ten minutes. Factor X activity was recorded as mOD/minute, and enzyme velocities over the linear portion of the reaction curve were compared to determine inhibition of factor X activation by the anti-TF antibodies.
- As shown in Figure 8, the CDR-grafted TF8-5G9 antibody is approximately as effective as the murine TF8-5G9 in inhibiting factor X activation. This indicates that the CDR-grafted TF8-5G9 is functionally active.

#### EXAMPLE 9

C nstruction of the CDR-Grafted Heavy and Light Chain Myeloma Expression Vectors

For the purpose of establishing a permanent 5 CDR-grafted antibody-producing cell line, the TF8HCDR20 and TF8LCDR3 genes were subcloned into myeloma cell expression vectors. The heavy chain TF8HCDR20 was subcloned into the EcoRI and BclI sites of the pEe6hCMV-BqlII myeloma expression vector described by Stephens et 10 al. (1989) Nucleic Acids Res. 17:7110 to produce pEe6TF8HCDR20. The light chain TF8LCDR3 was subcloned into the EcoTI and BclI sites of the pEel2 myeloma expression vector to produce pEel2TF8LCDR3. The heavy and light chain expression vectors are illustrated in 15 Figures 9 and 10, respectively. In both vectors antibody gene transcription was driven by the human cytomegalovirus (hCMV) promoter-enhancer, which lies directly 5' to the multiple cloning site. The polyadenylation signal sequence lies 3' to the multiple 20 cloning site and signals the termination of transcription. Each vector contains the ß-lactamase gene to allow for ampicillin selection in E. coli. The pEel2 vector contains a glutamine synthetase cDNA gene under the transcriptional control of the SV40 early 25 promoter. Glutamine synthetase allows for myeloma cell transfectants to be selected in glutamine-free media. Myeloma cells are devoid of glutamine synthetase activity and are dependent on a supply of glutamine in the culture media. Cells which have been transfected 30 with the pEel2 vector, containing the glutamine

WO 96/40921 PCT/US96/09287

-49-

synthetase gene, are able to synthesize glutamine from 1 glutamate and can survive in the absence of glutamine.

The pEe6TF8HCDR20 expression vector is a 7073 bp plasmid whose DNA sequence is shown in Figure 4 and SEQ ID NO:15. The coding regions of the TF8HCDR20 gene are 5 translated. The essential regions of this vector are described below:

- Nucleotides #1-2360: The TF8HCDR20 CDR-grafted HC gene is described in Example
   The HC gene was inserted as an EcoRI/BamHI fragment into the EcoRI/BclI sites of the pEe6hCMV-BqlII vector.
- 2. Nucleotides #2361-2593: This region encodes the SV40 early gene polyadenylation signal (SV40 nucleotides 2770-2537), which acts as a transcriptional terminator. This fragment is flanked by a 5' <u>Bcl</u>I site and a 3' <u>Bam</u>HI site. The 3' <u>Bam</u>HI end of the heavy chain gene was spliced to the 5' <u>Bcl</u>I site of the polyadenylation signal, thus eliminating both sites.

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- 3. Nucleotides #2594-3848: This region is a <a href="mailto:BamHI-Bql">BamHI-Bql</a>I fragment from pBR328
  (nucleotides 375-2422) but with a deletion between the SaI and AvaI sites (pBR328 nucleotides 651-1425) following the addition of a SalI linker to the AvaI site. This region contains the Col El bacterial origin of replication.
- 5. Nucleotides #4328-4885: This is an XmnI-HindIII fragment of the ColEl based plasmid pCT54 described by Emtage et al. (1983) Proc. Natl. Acad. Sci. USA

- 80:3671. The <u>Hind</u>III site was converted to a <u>Bql</u>II site by the addition of a linker following the addition of the hCMV promoter described below.
- 6. Nucleotides #4886-7022: These nucleotides encode the Pst-lm fragment of human cytomeglovirus (hCMV) strain AD 169 described by Greenway et al. (1982) Gene 18:355 containing the region coding for the hCMV middle intermediate early promoter. This Pst-lm fragment was cloned into the HindIII site of pEe6hCMV by addition of oligonucleotides of the following sequence to either end of the fragment:
  - 5' GTCACCGTCCTTGACACGA 3'
  - 3' ACGTCAGTGGCAGGAACTGTGCTTCGA 5'

The resulting 2100 bp fragment was inserted such that the promoter directed transcription towards the EcoRI site of pEe6hCMV. The oligonucleotide above served to recreate the complete 5' untranslated sequence of the hCMV-MIE gene the added irrelevant sequence at the very 5' end of the fragment. The HindIII site at the 5' end was subsequently converted to a BqlII site by the addition of a further linker.

- Nucleotides #7023-7073: The pSP64 polylinker with the <u>BamHI</u> and <u>Sa</u>II sites removed.
- 25 The pEel2TF8LCDR3 expression vector is a 7864 bp plasmid whose DNA sequence is shown in Figure 5 and SEQ ID NO:17. The coding regions of the TF8LCDR3 gene are translated. The essential regions of this expression vector are described below:
- Nucleotides #1-759: The TF8LCDR3 CDRgrafted LC gene is described in Example 7. The gene was inserted as an

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- EcoRI/BamHI fragment into the EcoRI/BclII sites of the pEel2 expression vector.
- Nucleotides #760-3284: These regions of pEe12 are identical to the regions encoded by nucleotides 2361-4885 of the pEe6TF8HCDR20 vector described above (regions #2-5).
- 3. Nucleotides #3285-5736: This region encodes the Chinese hamster ovary glutamine synthetase cDNA under the transcriptional control of the SV40 early promoter and followed by the SV40 polyadenylation and splice signals from 10 the pSV2.dhfr vector described by Subramani et al. (1981) Mol. Cell. Biol. 1:854. The following describes the derivation of this region: A 1200 bp Nael-Pvull fragment, containing a complete GS coding sequence, was excised from the Chinese hamster ovary cDNA clone  $\lambda$ GS1.1 described by Hayward <u>et al</u>. (1986) 15 Nucleic Acid Res. 14:999. After addition of a HindIII linker to the NaeI site and a BqlII linker to the PvuII site (hence destroying the NaeI and PvuII sites), the 1200 bp fragment was cloned in place of DHFR sequences in pSV2.dhfr between the HindIII and BqlII sites to form pSV2.GS. 20 The single remaining PvuII site in pSV2BamGS was converted to a BamHI site by addition of an oligonucleotide linker to form pSV2BamGS. An EcoRI site in the GS cDNA was destroyed by site directed mutagenesis without altering the amino acid sequence in pSV2BamGS and the HindIII site was destroyed by filling in 25 with DNa polymerase I. The 2451 bp BamHI fragment from this plasmid, containing the complete SV40-GS hybrid transcription unit, was excised and inserted at the BqlII site of pEe6hCMV-BqlII site of pEe6hCMV-BqlII such that transcription from the sV40 early promoter proceeds 30 towards the hCMV promoter.

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4. Nucleotides #5737-7864: This region is identical to the hCMV promoter and pSP64 polylinker encoded by nucleotides 4886-7073 of the pEe6TF8HCDR20 vector described above (regions 6 and 7).

For the purpose of ensuring that both the pEe6TF8HCDR20 and peE12TF8LCDR3 vectors co-transfected myeloma cells, the vectors were joined in linear concatamers. Both the pEe6TF8HCDR20 and pEe12TF8LCDR3 vectors were digested at the unique SalI site. The SalI linearized pEe6TF8HCDR20 vector was phosphatased at its 5' ends to prohibit ligation of two pEe6TF8HCDR20 vectors onto each other. This phosphatased HC vector was ligated in a 2:1 molar ratio to the Sal linearized pEe12TF8LCDR3. The resulting concatamers were most likely of the following composition:

15 SalI SalI SalI SalI

pEe6TF8HCDR20 pEe12TF8LCDR3 pEe6TF8HCDR20

This concatamerized DNA was extracted with phenol and chloroform, and precipitated with ammonium acetate and 20 ethanol. The DNA precipitate was resuspended in distilled water to a concentration of 1  $\mu$ g/ $\mu$ L and used to transfect myeloma cells.

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### EXAMPLE 10

Development of NSO Expression Cell Lines

Stably transformed cell lines expressing the humanized TF8-5G9 antibody were prepared by transfecting 5 CDR-grafted heavy and light chain expression vectors into NSO mouse myeloma cells. Selection of transfected cells was carried out using the dominant selectable marker gene, glutamine synthetase (GS).

The NSO mouse myeloma cell line, obtained from
10 Celltech, Ltd., is a subclone derived from NS-1 and does
not express intracellular light chains. These cells
were cultured in Dulbecco's modified Eagle's medium
(DMEM) with added glutamine and 10% fetal bovine serum
(FBS). To prepare for transfection, the cells were

15 harvested in mid-log phase of the growth cycle, centrifuged for 5 minutes, washed with phosphate buffered saline (PBS), centrifuged again, and the cell pellet was resuspended in 2.2 mL of PBS. The final cell concentration was 2.18 x 10 mL. Cells were maintained on ice during the entire procedure.

The DNA to be transfected (pEel2TF8LCDR3 x pEe6TF8HCDR20) was prepared as a concatamer as described in Example 9. The DNA and NSO cells were added to a 0.4 cm BioRad Gene Pulser cuvette in the following order:

 $\mu$ L (40  $\mu$ g) DNA concatamer  $\mu$ L double distilled water  $\mu$ L 10 x PBS  $\mu$ L NSO cells (8.72 x 10 $^6$  cells)

Transfection was performed by electroporation 30 following a protocol provided by Celltech, Ltd. In this procedure, the cells and DNA in PBS buffer were exposed to a brief, high voltage pulse of electricity causing l transient micropores to form on the cell membrane. DNA transfer takes place through these openings. To prepare for electroporation, the suspension of NSO cells and DNA was gently mixed and incubated on ice for 5 minutes.

- 5 The cuvette was placed in a BioRad Gene Pulser and given 2 consecutive electrical pulses at settings of 3  $\mu$ F (capacitance) and 1.5V (voltage). Following electroporation, the cuvette was returned to the ice for 5 minutes. The suspension was then diluted in prewarmed 10 growth medium and distributed into seven 96-well plates. Control plates containing cells electroporated without
  - control plates containing cells electroporated without DNA were also prepared at the same time to measure the presence of spontaneous mutants. Plates were placed in a 37°C incubator with 5% CO<sub>2</sub>.
- is an enzyme that converts glutamate to glutamine. NSO cells require glutamine for growth due to inadequate levels of endogenous GS gene expression. In the DNA concatamer, this gene is located on the pEel2TF8LCDR3 vector. Transfected cells which incorporate the GS gene become glutamine-independent. Cells not integrating the GS gene into their genome would remain glutamine-dependent and would not survive in glutamine-free medium. Approximately 18 hours post electroporation, all plates were fed with glutamine-free selection medium
- Approximately 3 weeks after transfection, distinct macroscopic colonies were observed. These were 30 screened for expression of the intact humanized antibody using the assembly ELISA as described in Example 5.

and returned to the incubator until viable colonies

appeared.

Tissue culture supernatants from wells containing
1 colonies were screened at a 1:10 dilution. Positive
wells showing activity greater than the 25 ng/mL
standard were subcultured and expanded for further
analysis.

For selection of high producers, antibody production was quantitated after a 96 hour growth period. Tissue culture flasks were seeded with 2 x  $10^5$  cells/mL in 10 mL of selection medium and incubated at  $37^{\circ}$ C,  $5^{\circ}$  CO<sub>2</sub> for 96 hours. At the end of that time 10 period, an aliquot was taken to determine cell concentration and antibody titer. Evaluation of antibody production was calculated as  $\mu$ g/mL and pg/cell/96 hours. The highest producers from this transfection were:

15	Cell Line	μq/mL	pg/cell/96 hour
	2B1	26.3	24.3
	3E11	27.6	<b>59.9</b> ·
•	4G6	30.2	41.9

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## EXAMPLE 11

CDR Grafted Antibody TF8HCDR20 x TF8LCDR3
Inhibits Tissue Factor In Vivo

CDR grafted antibody TF8HCDR20 x TF8LCDR3 was 5 compared to murine antibody TF8-5G9 for its ability to protect rats from experimentally induced disseminated intravascular coagulation (DIC). In the DIC model, rats are challenged with human thromboplastin (a crude tissue extract containing TF activity), resulting in fibrinogen consumption and death. Pretreatment of rats with anti-TF antibody was demonstrated to protect rats from fibrinogen consumption and death as follows.

Human thromboplastin was prepared as described in U.S. Patent 5,223,427. Saline control or 30  $\mu$ /ml of 15 TF8-5G9 or CDR-grafted antibody was injected through the tail vein of rats, followed by injection of thromboplastin equivalent to 200 ng of recombinant TF. Clotting times were determined at T=0 and T=1 minute as a measure of fibrinogen concentration. Clotting times 20 are proportional to fibrinogen concentration, with a 60 second clotting time corresponding to an 80% reduction in fibrinogen concentration. Clotting times of greater than 60 seconds cannot be accurately measured and were recorded as 60 seconds.

25 Survivability and clotting times for three representative studies are shown below.

		Survi	vors	•
	Study	Controls	TF8-5G9	CDR-grafted Ab
30	1	0/8	5/8	6/8
50	2	0/8	4/7	7/8
	3	0/8	8/8	3/7

1			<u>Clotting Times</u> <u>Controls</u>	-	
		y #1 <u>T=1</u>	Study #2 <u>T=0</u> <u>T=1</u>	Stud T=0	dy #3 <u>T=1</u>
5	16 16 17 15 16 16	>60 >60 >60 >60 >60 >60 >60 >60	18 >60 18 >60 18 >60 18 >60 16 >60 18 >60 17 >60 17 >60	19 21 18 19 18 18 18	>60 >60 >60 >60 >60 54 >60 >60 >60
10		-	Clotting Times Murine TF8-5G9		
	Stud T=0		Study #2 T=0 T=1	Stud T=0	y #3 <u>T=1</u>
15	16 15 15 15 16 16 16	36 41 33 31 >60 >60 33 33 >60	18 34 18 36 18 >60 17 >60 18 50 17 34 17 34 18 31	19 18 19 18 18 19 19	28 29 29 29 28 40 40 34 >60
20			Clotting Times		
	Stud T=0		CDR-grafted TF8-5G9 Study #2 T=0 T=1	Stud T=0	y #3 <u>T=1</u>
25	16 16 16 22 16 15	>60 >60 >60 37 32 >60 >60	17 >60 17 33 18 32 18 >60 17 32 18 31 17 31	21 18 17 20 17 18	>60 34 >60 35 58 33 31
20	16	>60	16 32		

WO 96/40921 PCT/US96/09287

-58-

Twenty-three of the twenty-four control rats

1 had clotting times of greater than 60 seconds indicating
that virtually all untreated rats were consuming more
than 80% of their fibrinogen. Both the CDR-grafted and
murine antibody treated rats had similar clotting times
5 at one minute of 44.5 and 40 seconds. Further, only six
of the murine antibody treated rats and nine of the CDRgrafted antibody treated rats had clotting times in
excess of 60 seconds. Accordingly, both the murine and
CDR-grafted antibodies were able to neutralize TF and
thus protect rats from fibrinogen consumption and death.

### SEQUENCE LISTING

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- (1) GENERAL INFORMATION:
  - (i) APPLICANT: Joliffe, Linda K. Zivin, Robert A. Pulito, Virginia L.

- (ii) TITLE OF INVENTION: CDR-GRAFTED ANTI-TISSUE FACTOR ANTIBODIES AND METHODS OF USE THEREOF
- (iii) NUMBER OF SEQUENCES: 20
- (iv) CORRESPONDENCE ADDRESS:
  - (A) ADDRESSEE: Scully, Scott, Murphy & Presser (B) STREET: 400 Garden City Plaza
- - (C) CITY: Garden City

    - (D) STATE: New York
      (E) COUNTRY: United States
    - (F) ZIP: 11530
  - (v) COMPUTER READABLE FORM:
    - (A) MEDIUM TYPE: Floppy disk
    - (B) COMPUTER: IBM PC compatible
- (C) OPERATING SYSTEM: PC-DOS/MS-DOS 15
  - (D) SOFTWARE: PatentIn Release #1.0, Version #1.25
  - (vi) CURRENT APPLICATION DATA:

    - (A) APPLICATION NUMBER: (B) FILING DATE: 07-JUN-1995
    - (C) CLASSIFICATION:
  - (viii)-ATTORNEY/AGENT INFORMATION:

    - (A) NAME: DiGiglio, Frank S. (B) REGISTRATION NUMBER: 31,346
    - (C) REFERENCE/DOCKET NUMBER: 9598
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1.		(i)	( Z ( E	A) LE B) TY C) ST	CE CE ENGTH (PE: TRANI OPOLO	i: 14 nucl	189 h leic ESS:	acio doul	pai:	rs							
5		•	FE!	ATURI A) NZ	LE TY E: AME/I OCATI	ŒY:	CDS	.,		<b>2)</b>				•		٠	,
10	GGT		ACA A	ATG A	CE DI	rgc <i>i</i>	AGC 1	rgg (	STC 1	ATC 1	TTC :						49
					AAT Asn												97
15	CTT Leu 30	GTG Val	AGG Arg	CCA Pro	GGG Gly	GCC Ala 35	TTA Leu	GTC Val	AAG Lys	TTG Leu	TCC Ser 40	TGC Cys	AAA Lys	GCT Ala	TCT Ser	GGC Gly 45	145
	TTC Phe	AAC Asn	ATT Ile	AAA Lys	GAC- Asp 50	TAC Tyr	TAT Tyr	ATG Met	CAC His	TGG Trp 55	GTG Val	AAG Lys	CAG Gln	AGG Arg	CCT Pro 60	GAA Glu	193
20	CAG Gln	GGC Gly	CTG Leu	GAG Glu 65	TGG Trp	ATT Ile	GGA Gly	TTG Leu	ATT Ile 70	GAT Asp	CCT Pro	GAG Glu	AAT Asn	GGT Gly 75	Asn	ACT Thr	241
20	ATA Ile	TAT Tyr	GAC Asp 80	CCG Pro	AAG Lys	TTC Phe	CAG Gln	GGC Gly 85	AAG Lys	GCC Ala	AGT Ser	ATA Ile	ACA Thr 90	GCA Ala	GAC Asp	ACA Thr	289
	TCC Ser	TCC Ser 95	AAC Asn	ACA Thr	GCC Ala	TAC Tyr	CTG Leu 100	CAG Gln	CTC Leu	AGC Ser	AGC Ser	CTG Leu 105	ACA Thr	TCT Ser	GAG Glu	GAC Asp	337
25	ACT Thr 110	GCC	GTC Val	TAT Tyr	TAC Tyr	TGT Cys 115	GCT Ala	AGA Arg	GAT Asp	AAC Asn	TCG Ser 120	TAC Tyr	TAC Tyr	TTT Phe	GAC Asp	TAC Tyr 125	385

1							GCC Ala			433
							GCC Ala			481
5							TTC Phe			529
			-				GGT Gly 185			577
10							AGC Ser			625
			 				ACC Thr			673
							ATT Ile			721
15							GAA Glu			769
		Phe					ACC Thr 265			817
20	Lys						AAG Lys			865
							GTG Val			913
25							TTC Phe			961

1					ATG Met												1009
					AGT Ser												1057
5					GGC Gly												1105
					CAG Gln 370												1153
10					TTC Phe												1201
					GAG Glu												1249
					TTC Phe												1297
15					AAT Asn												1345
					ACT Thr 450											T	1391
20	GAT	CCA	STG :	rcct:	rggao	GC C	CTCT	GTC	C TAC	CAGG	ACTC	TGA	CACC	CAC C	CTCC	ACCCCT	1451
	CCC:	rgta:	raa 1	ATAA	AGCAG	CC C1	AGCA	CTGC	TTC	GAC	CC						1489

## (2) INFORMATION FOR SEQ ID NO:2:

(i) SEQUENCE CHARACTERISTICS:

(A) LENGTH: 460 amino acids
(B) TYPE: amino acid
(D) TOPOLOGY: linear

(ii)	MOLECULE	TYPE:	protein
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(xi) SEQUENCE DESCRIPTION: SEQ ID NO:2:

Met Lys Cys Ser Trp Val Ile Phe Phe Leu Met Ala Val Val Thr Gly
1 5 10 15

Val Asn Ser Glu Ile Gln Leu Gln Gln Ser Gly Ala Glu Leu Val Arg 5 20 25 30

Pro Gly Ala Leu Val Lys Leu Ser Cys Lys Ala Ser Gly Phe Asn Ile 35 45

Lys Asp Tyr Tyr Met His Trp Val Lys Gln Arg Pro Glu Gln Gly Leu 50 60

Glu Trp Ile Gly Leu Ile Asp Pro Glu Asn Gly Asn Thr Ile Tyr Asp 10  $\phantom{0}65\phantom{0}$  70  $\phantom{0}75\phantom{0}$  80

Pro Lys Phe Gln Gly Lys Ala Ser Ile Thr Ala Asp Thr Ser Ser Asn 85 90 95

Thr Ala Tyr Leu Gln Leu Ser Ser Leu Thr Ser Glu Asp Thr Ala Val 100 105 110

Tyr Tyr Cys Ala Arg Asp Asn Ser Tyr Tyr Phe Asp Tyr Trp Gly Gln 15  $\sim$  115 120 125

Gly Thr Thr Leu Thr Val Ser Ser Ala Lys Thr Thr Pro Pro Ser Val 130 135 140

Tyr Pro Leu Ala Pro Gly Ser Ala Ala Gln Thr Asn Ser Met Val Thr 145 150 155 160

Leu Gly Cys Leu Val Lys Gly Tyr Phe Pro Glu Pro Val Thr Val Thr 20 165 170 175

Trp Asn Ser Gly Ser Leu Ser Ser Gly Val His Thr Phe Pro Ala Val 180 185 190

Leu Gln Ser Asp Leu Tyr Thr Leu Ser Ser Ser Val Thr Val Pro Ser

Ser Thr Trp Pro Ser Glu Thr Val Thr Cys Asn Val Ala His Pro Ala 25 210 215 220

30

1	Ser 225	Ser	Thr	Lys	Val	Asp 230	Lys	Lys	Ile	Val	Pro 235	Arg	Asp	Сув	Gly	Сув 240
4	ГÀв	Pro	Cys	Ile	Cys 245	Thr	Val	Pro	Glu	Val 250	Ser	Ser	Val	Phe	Ile 255	Phe
	Pro	Pro	Lys	Pro 260	Lys	Asp	Val	Leu	Thr 265	Ile	Thr	Leu	Thr	Pro 270	Lys	Val
5	Thr	Сув	Val 275	Val	Val	qaA	Ile	Ser 280	Lys	Asp	Asp	Pro	Glu 285	Val	Gln	Ph∈
	Ser	Trp 290	Phe	Val	Asp	Asp	Val 295	Glu	Val	His	Thr	Ala 300	Gln	Thr	Gln	Pro
	Arg 305	Glu	Glu	Gln	Phe	Asn 310	Ser	Thr	Phe	Arg	Ser 315	Val	Ser	Glu	Leu	Pro 320
10	Ile	Met	His	Gln	Asp 325	Trp	Leu	Asn	Gly	330 Lys	Glu	Phe	Lys	Сув	Arg 335	Val
	Asn	Ser	Ala	Ala 340	Phe	Pro	Ala	Pro	Ile 345	Glu	Lys	Thr	Ile	Ser 350	ГÀа	Thr
	Lув	Gly	Arg 355	Pro	Lys	Ala	Pro	Gln 360	Val	Tyr	Thr	Ile	Pro 365	Pro	Pro	Lys
15	Glu	Gln 370	Met	Ala	Lys	Asp	Lys 375	Val	Ser	Leu	Asn	380 Cys	Met	Ile	Thr	Asg
	Phe 385	Phe	Pro	Glu	Yab	Ile 390	Thr	Val	Glu	Trp	Gln 395	Trp	Asn	Gly	Gln	Pro 400
	Ala	Glu	Asn	Tyr	Lys 405	Asn	Thr	Gln	Pro	Ile 410	Met	Asp	Thr	Asp	Gly 415	Ser
20	Tyr	Phe	Val	Tyr 420	Ser	Lys	Leu	Asn	Val 425	Gln	Lys	Ser	Asn	Trp 430	Glu	Ala
	Gly	Asn	Thr 435	Phe	Thr	Сув	Ser	Val 440	Leu	His	Glu	Gly	Leu 445	His	Asn	His
	His	Thr 450	Glu	Lys	Ser	Leu	Ser 455	His	Ser	Pro	Gly	Lys 460				

25

	(2)	INFO	ORMA!	rion	FOR	SEQ	ID 1	10:3	:								
1		(i)	(1 (1	QUENCA) LI B) TY C) SY D) TO	INGTI (PE: [RANI	nuc nuc DEDNI	37 ba Leic ESS:	ase p acid doub	pair: d	5							•
5			) FE2	LECUI ATURI A) NI B) LO	e: Ame/1	KEY:	CDS										
10	GGA	ATC Met	CGC	QUENC G GCC G Ala	c cci	r GC:	CAC	G TT	r TT:	r GGO	ATC	e Lev	F TTO	CTO	C TG( 1 Tr)	G TTT Phe 15	49
	CCA Pro	GGT Gly	ATC Ile	AGA Arg	TGT Cys 20	GAC Asp	ATC Ile	AAG Lys	ATG Met	ACC Thr 25	CAG Gln	TCT Ser	CCA Pro	TCC Ser	TCC Ser 30	ATG Met	97
15	TAT	GCA Ala	TCG Ser	CTG Leu 35	GGA Gly	GAG Glu	AGA Arg	GTC Val	ACT Thr 40	ATC Ile	ACT Thr	TGT Cys	AAG Lys	GCG Ala 45	AGT Ser	CAG Gln	145
	GAC Asp	ATT Ile	AGA Arg 50	AAG Lys	TAT Tyr	TTA Leu	AAC Asn	TGG Trp 55	TAC	CAG Gln	CAG Gln	AAA Lys	CCA Pro 60	TGG Trp	AAA Lys	TCT Ser	193
20	CCT Pro	AAG Lys 65	ACC Thr	CTG Leu	ATC	TAT Tyr	TAT Tyr 70	GCA Ala	ACA Thr	AGC Ser	TTG Leu	GCA Ala 75	GAT Asp	GGG Gly	GTC Val	CCA Pro	241
20	TCA Ser 80	AGA Arg	TTC Phe	AGT Ser	GC	AGT Ser 85	GGA Gly	TCT Ser	GGG Gly	CAA Gln	GAT Asp 90	TAT Tyr	TCT Ser	CTA Leu	ACC Thr	ATC Ile 95	289
	AGC Ser	AGC Ser	CTG Leu	GAG Glu	TCT Ser 100	GAC Asp	GAT Asp	ACA Thr	GCA Ala	ACT Thr 105	TAT Tyr	TAC Tyr	TGT Cys	CTA Leu	CAA Gln 110	His	337
25	GGT Gly	GAG Glu	AGC Ser	CCG Pro 115	TAC Tyr	ACG Thr	TTC Phe	GGA Gly	GGG Gly 120	gly ggg	ACC Thr	AAG Lys	CTG Leu	GAA Glu 125	ATA Ile	AAC Asn	385

30 '

1															AGT Ser		4	133
															AAC Asn		4	81
5															GAA Glu		5	529
															GAC Asp 190		5	577
10															TAT Tyr		6	525
															ACT Thr		6	573
								AAG Lys				TAG	AGAC	AAA (	GGTC	CTGAGA		726
15	CGC	CACC	ACC Z	AGCT	ccc	AG C	rcca:	CCT	A TC	TCC	CTTC	TAAC	GTC	rtg (	GAGG	CTTCCC	7	786
	CAC	AAGC	GAC (	CTAC	CACT	ST T	cccc:	IGCT	CA	AACC:	CCT	ccc	CACC	rcc :	TTCT	CCTCCT	ε	346
	CCT	CCCT	TTC (	CTTG	GCTT:	TT A	CAT	GCTA	A TA	TTTG	CAGA	AAA:	TATT	CAA :	TAAAC	STGAGT	. 9	906
	CTT	rgca(	cii (	GAAA	AAAA	AA A	AAAA	AAAA	A A				•				9	937
20		•								•								

## (2) INFORMATION FOR SEQ ID NO:4:

- (i) SEQUENCE CHARACTERISTICS:
  (A) LENGTH: 234 amino acids
  (B) TYPE: amino acid
  (D) TOPOLOGY: linear
- 25
- (ii) MOLECULE TYPE: protein

### (xi) SEQUENCE DESCRIPTION: SEQ ID NO:4:

- 1 Met Arg Ala Pro Ala Gln Phe Phe Gly Ile Leu Leu Leu Trp Phe Pro 1 15
  - Gly Ile Arg Cys Asp Ile Lys Met Thr Gln Ser Pro Ser Ser Met Tyr
    20 25 30
- Ala Ser Leu Gly Glu Arg Val. Thr Ile Thr Cys Lys Ala Ser Gln Asp 35 40
  - Ile Arg Lys Tyr Leu Asn Trp Tyr Gln Gln Lys Pro Trp Lys Ser Pro 50 60
  - Lys Thr Leu Ile Tyr Tyr Ala Thr Ser Leu Ala Asp Gly Val Pro Ser 65 70 75 80
- Arg Phe Ser Gly Ser Gly Ser Gly Gln Asp Tyr Ser Leu Thr Ile Ser 10 85 90 95
  - Ser Leu Glu Ser Asp Asp Thr Ala Thr Tyr Tyr Cys Leu Gln His Gly 100 105 110
    - Glu Ser Pro Tyr Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Asn Arg
- Ala Asp Ala Ala Pro Thr Val Ser Ile Phe Pro Pro Ser Ser Glu Gln 15 130 140
  - Leu Thr Ser Gly Gly Ala Ser Val Val Cys Phe Leu Asn Asn Phe Tyr 145 150 155 160
  - Pro Lys Asp Ile Asn Val Lys Trp Lys Ile Asp Gly Ser Glu Arg Gln 165 170 175
- Asn Gly Val Leu Asn Ser Trp Thr Asp Gln Asp Ser Lys Asp Ser Thr 20 180 185 190
  - Tyr Ser Met Ser Ser Thr Leu Thr Leu Thr Lys Asp Glu Tyr Glu Arg
  - His Asn Ser Tyr Thr Cys Glu Ala Thr His Lys Thr Ser Thr Ser Pro 210 215 220
- Asn Val Lys Ser Phe Asn Lys Asn Glu Cys 25 225 230

```
(2) INFORMATION FOR SEQ ID NO:5:
```

1 (i) SEQUENCE CHARACTERISTICS:

- (A) LENGTH: 5 amino acids
- (B) TYPE: amino acid
- (C) STRANDEDNESS: double
- (D) TOPOLOGY: linear
- (ii) MOLECULE TYPE: peptide

5

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:5:

Asp Asp Tyr Met His

# 10 (2) INFORMATION FOR SEQ ID NO:6:

- (i) SEQUENCE CHARACTERISTICS:
  - (A) LENGTH: 17 amino acids
  - (B) TYPE: amino acid
  - (C) STRANDEDNESS: double
  - (D) TOPOLOGY: linear
- (ii) MOLECULE TYPE: peptide

15

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:6:

Leu Ile Asp Pro Glu Asn Gly Asn Thr Ile Tyr Lys Pro Lys Phe Gln 1 5 10

Gly

20

- (2) INFORMATION FOR SEQ ID NO:7:
  - (i) SEQUENCE CHARACTERISTICS:
    - (A) LENGTH: 8 amino acids
    - (B) TYPE: amino acid
    - (C) STRANDEDNESS: double
  - (D) TOPOLOGY: linear

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(ii) MOLECULE TYPE: peptide

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- (xi) SEQUENCE DESCRIPTION: SEQ ID NO:7:
- 1 Asp Asn Ser Tyr Tyr Phe Asp Tyr
- (2) INFORMATION FOR SEQ ID NO:8:
- (i) SEQUENCE CHARACTERISTICS:
  (A) LENGTH: 11 amino acids 5

  - (B) TYPE: amino acid (C) STRANDEDNESS: double (D) TOPOLOGY: linear

  - (ii) MOLECULE TYPE: peptide
- (xi) SEQUENCE DESCRIPTION: SEQ ID NO:8: 10 Lys Ala Ser Gln Asp Ile Arg Lys Tyr Leu Asn
  - (2) INFORMATION FOR SEQ ID NO:9:
- 15 (i) SEQUENCE CHARACTERISTICS:
  - (A) LENGTH: 7 amino acids

    - (B) TYPE: amino acid (C) STRANDEDNESS: double
    - (D) TOPOLOGY: linear
  - (ii) MOLECULE TYPE: peptide
- (xi) SEQUENCE DESCRIPTION: SEQ ID NO:9: 20 Tyr Ala Thr Ser Leu Ala Asp

25

(2) INFORMATION FOR SEC ID NO:	(2)	INFORMATION	FOR	SEO	ID	NO:10
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l (i) SEQUENCE CHARACTERISTICS:

- (A) LENGTH: 9 amino acids
- (B) TYPE: amino acid
- (C) STRANDEDNESS: double
- (D) TOPOLOGY: linear

(ii) MOLECULE TYPE: peptide

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(xi) SEQUENCE DESCRIPTION: SEQ ID NO:10:

Leu Gln His Gly Glu Ser Pro Tyr Thr

### 10 (2) INFORMATION FOR SEQ ID NO:11:

- (i) SEQUENCE CHARACTERISTICS:
  - (A) LENGTH: 117 amino acids
  - (B) TYPE: amino acid
  - (C) STRANDEDNESS: single
  - (D) TOPOLOGY: linear
- (ii) MOLECULE TYPE: peptide

15

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:11:

Gln Val Gln Leu Val Gln Ser Gly Gly Gly Val Val Gln Pro Gly Arg
1 5 10 15

Leu Leu Arg Leu Ser Cys Lys Ala Ser Gly Phe Asn Ile Lys Asp Tyr 20 25 0

Tyr Met His Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Ile

Gly Leu Ile Asp Pro Glu Asn Gly Asn Thr Ile Tyr Asp Pro Lys Phe 50 60

Gln Gly Arg Phe Ser Ile Ser Ala Asp Thr Ser Lys Asn Thr Ala Phe 65 70 75 80

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Leu Gln Met Asp Ser Leu Arg Pro Glu Asp Thr Ala Val Tyr Tyr Cys 95

Ala Arg Asp Asn Ser Tyr Tyr Phe Asp Tyr Trp Gly Gln Gly Thr Pro 100

Val Thr Val Ser Ser 115

5

- (2) INFORMATION FOR SEQ ID NO:12:
  - (i) SEQUENCE CHARACTERISTICS:
    - (A) LENGTH: 108 amino acids
    - (B) TYPE: amino acid
    - (C) STRANDEDNESS: single
    - (D) TOPOLOGY: linear

10

- (ii) MOLECULE TYPE: peptide
- (xi) SEQUENCE DESCRIPTION: SEQ ID NO:12:

Asp Ile Gln Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly
1 10 15

Asp Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Asp Ile Arg Lys Tyr 20 25 30

Leu Asn Trp Tyr Gln Gln Lys Pro Trp Lys Ala Pro Lys Thr Leu Ile 35

Tyr Ala Thr Ser Leu Ala Asp Gly Val Pro Ser Arg Phe Ser Gly 50 60

Ser Gly Ser Gly Thr Asp Tyr Thr Phe Thr Ile Ser Ser Leu Gln Pro 65 70 75

Glu Asp Ile Ala Thr Tyr Tyr Cys Leu Gln His Gly Glu Ser Pro Tyr 85 90 95

Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile Thr Arg

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(2)	) :	INFORMATIC	N FOR	SEQ	ID	NO:	13	3 :
-----	-----	------------	-------	-----	----	-----	----	-----

- 1 (i) SEQUENCE CHARACTERISTICS:
  - (A) LENGTH: 117 amino acids
  - (B) TYPE: amino acid
  - (C) STRANDEDNESS: single
  - (D) TOPOLOGY: linear
  - (ii) MOLECULE TYPE: peptide

- (xi) SEQUENCE DESCRIPTION: SEQ ID NO:13:
- Gln Val Gln Leu Val Glu Ser Gly Gly Gly Val Val Gln Pro Gly Arg 1 10 15

Ser Leu Arg Leu Ser Cys Lys Ala Ser Gly Phe Asn Ile Lys Asp Tyr 20 25 30

10

Tyr Met His Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Ile 35 40 45

Gly Leu Ile Asp Pro Glu Asn Gly Asn Thr Ile Tyr Asp Pro Lys Phe 50 60

Gln Gly Arg Phe Thr Ile Ser Ala Asp Asn Ser Lys Asn Thr Leu Phe 65 70 75 80

Leu Gln Met Asp Ser Leu Arg Pro Glu Asp Thr Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Asp Asn Ser Tyr Tyr Phe Asp Tyr Trp Gly Gln Gly Thr Pro 100 105 110

Val Thr Val Ser Ser 115

20

### (2) INFORMATION FOR SEQ ID NO:14:

- (i) SEQUENCE CHARACTERISTICS:
  - (A) LENGTH: 108 amino acids
  - (B) TYPE: amino acid
  - (C) STRANDEDNESS: single
- 25 (D) TOPOLOGY: linear

30

(ii) MOLECULE TYPE: peptide

1 (xi) SEQUENCE DESCRIPTION: SEQ ID NO:14: Asp Ile Gln Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly Asp Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Asp Ile Arg Lys Tyr 5 Leu Asn Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile Tyr Tyr Ala Thr Ser Leu Ala Asp Gly Val Pro Ser Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Tyr Thr Phe Thr Ile Ser Ser Leu Gln Pro 10 Glu Asp Ile Ala Thr Tyr Tyr Cys Leu Gln His Gly Glu Ser Pro Tyr Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile Thr Arg

- (2) INFORMATION FOR SEQ ID NO:15:
  - (i) SEQUENCE CHARACTERISTICS:
    - (A) LENGTH: 7073 base pairs
      (B) TYPE: nucleic acid
      (C) STRANDEDNESS: double

    - (D) TOPOLOGY: linear
- (ii) MOLECULE TYPE: peptide 20
  - (ix) FEATURE:

    - (A) NAME/KEY: CDS
      (B) LOCATION: 61..717
  - (ix) FEATURE:
    - (A) NAME/KEY: CDS
    - (B) LOCATION: 1111..1146

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(ix) FEATURE:

1				A) NI B) L				31	594									
		(ix	· (2	ATURI A) NI B) LO	AME/I				012			-					•	
5		(xi	) SE	QUENC	CE DI	escr:	IPTIC	ON: S	SEQ :	ID NO	0:15	:						
	GAA:	rtcg	CCT (	CCAC	CATGO	GA A	rggao	CTG	G GT	CTTT	CTCT	TCT	CTT	GTC	AGTA	ACTACA		61
	GGT Gly 1	GTA Val	CAC His	TCA Ser	CAA Gln 5	GTT Val	CAG Gln	CTG Leu	GTG Val	GAG Glu 10	TCT Ser	GGA Gly	GGA Gly	GGA Gly	GTA Val 15	GTA Val	10	) ł
10	CAA Gln	CCT Pro	GGA Gly	AGG Arg 20	TCA Ser	CTG Leu	AGA Arg	CTG Leu	TCT Ser 25	TGT Cys	AAG Lys	GCT Ala	AGT Ser	GGA Gly 30	TTC Phe	AAT Asn	15	56
	ATC Ile	AAG Lys	GAC Asp 35	TAT Tyr	TAT Tyr	ATG Met	CAC His	TGG Trp 40	GTC Val	AGA Arg	CAA Gln	GCT Ala	CCT Pro 45	GGA Gly	AAA Lys	GGA Gly	20	)4
15	CTC Leu	GAG Glu 50	TGG Trp	ATA Ile	GGT Gly	TTA Leu	ATT Ile 55	GAT Asp	CCT Pro	GAG Glu	TAA neA	GGT Gly 60	AAC Asn	ACG Thr	ATA Ile	TAT Tyr	25	52
	GAT Asp 65	CCC Pro	AAG Lys	TTC Phe	CAA Gln	GGA Gly 70	AGA Arg	TTC Phe	ATA Ile	ATT	TCT Ser 75	GCA Ala	GAC Asp	AAC Asn	TCT Ser	AAG Lys 80	30	)(
20	AAT Asn	ACA Thr	CTG Leu	TTC Phe	CTG Leu 85	CAG Gln	ATG Met	GAC Asp	TCA Ser	CTC Leu 90	AGA Arg	CCT Pro	GAG Glu	GAT Asp	ACA Thr 95	GCA Ala	34	18
20	GTC Val	TAC Tyr	TTT Phe	TGT Cys 100	GCT Ala	AGA Arg	GAT Asp	AAC Asn	AGT Ser 105	TAT Tyr	TAC Tyr	TTC Phe	GAC Asp	TAC Tyr 110	TGG Trp	GGC Gly	39	)€
	CAA Gln	GGA Gly	ACA Thr 115	CCA Pro	GTC Val	ACC Thr	GTG Val	AGC Ser 120	TCA Ser	GCT Ala	TCC Ser	ACC Thr	AAG Lys 125	GGC Gly	CCA Pro	TCC Ser	44	14
25	GTC Val	TTC Phe 130	CCC Pro	CTG Leu	GCG Ala	CCC Pro	TGC Cys 135	TCC Ser	AGG Arg	AGC Ser	ACC Thr	TCC Ser 140	GAG Glu	AGC Ser	ACA Thr	GCC Ala	49	)2

	GCC CTG GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG Ala Leu Gly Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val	540
1	145 150 155 160	,
	TCG TGG AAC TCA GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT Ser Trp Asn Ser Gly Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala	588
	165 170 175	
5	GTC CTA CAG TCC TCA GGA CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG Val Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val	636
כ	180 185 190	
	CCC TCC AGC AGC TTG GGC ACG AAG ACC TAC ACC TGC AAC GTA GAT CAC	684
	Pro Ser Ser Leu Gly Thr Lys Thr Tyr Thr Cys Asn Val Asp His 195 200 205	
	ANG CCC AGC ANC ACC ANG GTG GAC ANG AGA GTT GGTGAGAGGC CAGCACAGGG	737
10	Lys Pro Ser Asn Thr Lys Val Asp Lys Arg Val 210 215	
	CAGGGAGGGT GTCTGCTGGA AGCCAGGCTC AGCCCTCCTG CCTGGACGCA CCCCGGCTGT	797
	GCAGCCCCAG CCCAGGGCAG CAAGGCATGC CCCATCTGTC TCCTCACCCG GAGGCCTCTG	857
	ACCACCCCAC TCATGCTCAG GGAGAGGGTC TTCTGGATTT TTCCACCAGG CTCCGGGCAG	917
	CCACAGGCTG GATGCCCCTA CCCCAGGCCC TGCGCATACA GGGGCAGGTG CTGCGCTCAG	977
15	ACCTGCCAAG AGCCATATCC GGGAGGACCC TGCCCCTGAC CTAAGCCCAC CCCAAAGGCC	1037
	AAACTCTCCA CTCCCTCAGC TCAGACACCT TCTCTCCTCC CAGATTCGAG TAACTCCCAA	1097
	TCTTCTCTCT GCA GAG TCC AAA TAT GGT CCC CCA TGC CCA TCA TGC CCA	1146
	Glu Ser Lys Tyr Gly Pro Pro Cys Pro Ser Cys Pro 1 5 10	
20	GGTAAGCCAA CCCAGGCCTC GCCCTCCAGC TCAAGGCGGG ACAGGTGCCC TAGAGTAGCC	1206
	TGCATCCAGG GACAGGCCCC AGCCGGGTGC TGACGCATCC ACCTCCATCT CTTCCTCAGC	1266
	A CCT GAG TTC CTG GGG GGA CCA TCA GTC TTC CTG TTC CCC CCA AAA	1312
	Pro Glu Phe Leu Gly Gly Pro Ser Val Phe Leu Phe Pro Pro Lys 1 5 10 15	•
	CCC AAG GAC ACT CTC ATG ATC TCC CGG ACC CCT GAG GTC ACG TGC GTG	1360
25	Pro Lys Asp Thr Leu Met Ile Ser Arg Thr Pro Glu Val Thr Cys Val 20 25 30	

1	GTG Val	GTG Val	GAC Asp	GTG Val 35	AGC Ser	CAG Gln	GAA Glu	GAC Asp	CCC Pro 40	GAG Glu	GTC Val	CAG Gln	TTC Phe	AAC Asn 45	TGG Trp	TAC Tyr	1408
												AAG Lys					1456
5								Val.				CTC Leu 75					1504
												AAG Lys					1552
10												TAa YYY					1594
	GGT	GGA	CCC 1	ACGG	GTG	CG AC	GGC	CACAT	r GG2	ACAG	AGGT	CAG	CTCG	SCC (	CACC	CTCTGC	1654
	CCT	GGA(	STG 1	ACCG	CTGTC	GC CI	AACC	rctg:	CCC	CTAC	_				_	G CCA L Pro	1709
15												ATG Met					1757
						-						CCC					1805
20			_									AAC Asn 50					1853
												CTC Leu					1901
05												GTC Val					1949

1	GTG ATG CAT GAG GCT CTG CAC AAC CAC TAC ACA CAG AAG AGC CTC TCC Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Lys Ser Leu Ser 90 95 100	1997
	CTG TCT CTG GGT AAA TGAGTGCCAG GGCCGGCAAG CCCCCGCTCC CCGGGCTCTC Leu Ser Leu Gly Lys 105	2052
5	GGGGTCGCGC GAGGATGCTT GGCACGTACC CCGTCTACAT ACTTCCCAGG CACCCAGCAT	2112
٠,	GGAAATAAAG CACCCACCAC TGCCCTGGGGC CCCTGTGAGA CTGTGATGGT TCTTTCCACG	2172
	GGTCAGGCCG AGTCTGAGGC CTGAGTGACA TGAGGGAGGC AGAGCGGGTC CCACTGTCCC	2232
	CACACTGGCC CAGGCTGTGC AGGTGTGCCT GGGCCACCTA GGGTGGGGCT CAGCCAGGGG	2292
	CTGCCCTCGG CAGGGTGGGG GATTTGCCAG CGTGGCCCTC CCTCCAGCAG CAGGACTCTA	2352
10	GAGGATCATA ATCAGCCATA CCACATTTGT AGAGGTTTTA CTTGCTTTAA AAAACCTCCC	2412
	ACACCTCCCC CTGAACCTGA AACATAAAAT GAATGCAATT GTTGTTGTTA ACTTGTTTAT	2472
	TGCAGCTTAT AATGGTTACA AATAAAGCAA TAGCATCACA AATTTCACAA ATAAAGCATT	2532
	TTTTTCACTG CATTCTAGTT GTGGTTTGTC CAAACTCATC AATGTATCTT ATCATGTCTG	2592
1 =	GATECTETAC GCCGGACGCA TCGTGGCCGG CATCACCGGC GCCACAGGTG CGGTTGCTGG	2652
בי	CGCCTATATC GCCGACATCA CCGATGGGGA AGATCGGGCT CGCCACTTCG GGCTCATGAG	2712
	CGCTTGTTTC GGCGTGGGTA TGGTGGCAGG CCCGTGGCCG GGGGACTGTT GGGCGCCATC	2772
	TCCTTGCATG CACCATTCCT TGCGGCGGGG GTGCTCAACG GCCTCAACCT ACTACTGGGC	2832
	TGCTTCCTAA TGCAGGAGTC GCATAAGGGA GAGCGTCGAC CTCGGGCCGC GTTGCTGGCG	2892
20	TTTTTCCATA GGCTCCGCCC CCCTGACGAG CATCACAAAA ATCGACGCTC AAGTCAGAGG	2952
	TGGCGAAACC CGACAGGACT ATAAAGATAC CAGGCGTTTC CCCCTGGAAG CTCCCTCGTG	3012
	CGCTCTCCTG TTCCGACCCT GCCGCTTACC GGATACCTGT CCGCCTTTCT CCCTTCGGGA	3072
	AGCGTGGCGC TTTCTCAATG CTCACGCTGT AGGTATCTCA GTTCGGTGTA GGTCGTTCGC	3132
	TCCAAGCTGG GCTGTGTGCA CGAACCCCCC GTTCAGCCCG ACCGCTGCGC CTTATCCGGT	3192
25	AACTATCGTC TTGAGTCCAA CCCGGTAAGA CACGACTTAT CGCCACTGGC AGCAGCCACT	3252

	GGTAACAGGA	TTAGCAGAGC	GAGGTATGTA	GGCGGTGCTA	CAGAGTTCTT	GAAGTGGTGG	3312
1	CCTAACTACG	GCTACACTAG	AAGGACAGTA	TTTGGTATCT	GCGCTCTGCT	GAAGCCAGTT	3372
	ACCTTCGGAA	AAAGAGTTGG	TAGCTCTTGA	TCCGGCAAAC	AAACCACCGC	TGGTAGCGGT	3432
	GGTTTTTTTG	TTTGCAAGCA	GCAGATTACG	CGCAGAAAAA	AAGGATCTCA	AGAAGATCCT	3492
5	TTGATCTTTT	CTACGGGGTC	TGACGCTCAG	TGGAACGAAA	ACTCACGTTA	AGGGATTTTG	3552
)	GTCATGAGAT	TATCAAAAAG	GATCTTCACC	TAGATCCTTT	TAAATTAAAA	ATGAAGTTTT	3612
	AAATCAATCT	AAAGTATATA	TGAGTAAACT	TGGTCTGACA	GTTACCAATG	CTTAATCAGT	3672
	GAGGCACCTA	TCTCAGCGAT	CTGTCTATTT	CGTTCATCCA	TAGTTGCCTG	ACTCCCCGTC	3732
	GTGTAGATAA	CTACGATACG	GGAGGGCTTA	CCATCTGGCC	CCAGTGCTGC	AATGATACCG	3792
10	CGAGACCCAC	GCTCACCGGC	TCCAGATTTA	TCAGCAATAA	ACCAGCCAGC	CGGAAGGGCC	3852
	GAGCGCAGAA	GTGGTCCTGC	AACTTTATCC	GCCTCCATCC	AGTCTATTAA	TTGTTGCCGG	3912
	GAAGCTAGAG	TAAGTAGTTC	GCCAGTTAAT	AGTTTGCGCA	ACGTTGTTGC	CATTGCTACA	3972
	GGCATCGTGG	TGTCACGCTC	GTCGTTTGGT	ATGGCATCAT	TCAGCTCCGG	TTCCCAACGA	4032
15	TCAAGGCGAG	TTACATGATC	CCCCATGTTG	TGCAAAAAAG	CGGTTAGCTC	CTTCGGTCCT	4092
יי	CCGATCGTTG	TCAGAAGTAA	GTTGGCCGCA	GTGTTATCAC	TCATGGTTAT	GGCAGCACTG	4152
	CATAATTCTC	TTACTGTCAT	GCCATCCGTA	AGATGCTTTT	CTGTGACTGG	TGAGTACTCA	4212
•	ACCAAGTCAT	TCTGAGAATA	GTGTATGCGG	CGACCGAGTT	GCTCTTGCCC	GGCGTCAACA	4272
	CGGGATAATA	CCGCGCCACA	TAGCAGAACT	TTAAAAGTGC	TCATCATTGG	AAAACGTTCT	4332
20	TCGGGGCGAA	AACTCTCAAG	GATCTTACCG	CTGTTGAGAT	CCAGTTCGAT	GTAACCCACT	4392
	CGTGCACCCA	ACTGATCTTC	AGCATCTTTT	ACTITCACCA	GCGTTTCTGG	GTGAGCAAAA	4452
	ACAGGAAGGC	AAAATGCCGC	AAAAAAGGGA	ATAAGGGCGA	CACGGAAATG	TTGAATACTC	4512
	ATACTCTTCC	TTTTTCAATA	TTATTGAAGC	ATTTATCAGG	GTTATTGTCT	CATGAGCGGA	4572
25	TACATATTTG	AATGTATTTA	GAAAAATAAA	CAAATAGGGG	TTCCGCGCAC	ATTTCCCCGA	4632
- )	AAAGTGCCAC	CTGACGTCTA	AGAAACCATT	ATTATCATGA	CATTAACCTA	TAAAAATAGG	4692

	CGTATCACGA	GGCCCTGATG	GCTCTTTGCG	GCACCCATCG	TTCGTAATGT	TCCGTGGCAC	4752
1	CGACGACAAC	CCTCAAGAGA	AAATGTAATC	ACACTGGCTC	ACCTTCGGGT	GGGCCTTTCT	4812
	GCGTTTATAA	GGAGACACTT	TATGTTTAAG	AAGGTTGGTA	AATTCCTTGC	GGCTTTGGCA	4872
	GCCAAGCTAG	AGATCTCTAG	CTTCGTGTCA	AGGACGGTGA	CTGCAGTGAA	TAATAAAATG	4932
5	TGTGTTTGTC	CGAAATACGC	GTTTTGAGAT	TTCTGTCGCC	GACTAAATTC	ATGTCGCGCG	4992
כ	ATAGTGGTGT	TTATCGCCGA	TAGAGATGGC	GATATTGGAA	AAATCGATAT	TTGAAAATAT	5052
	GGCATATTGA	AAATGTCGCC	GATGTGAGTT	TCTGTGTAAC	TGATATCGCC	ATTTTTCCAA	5,112
	AAGTGATTTT	TGGGCATACG	CGÁTATCTGG	CGATAGCGCT	TATATCGTTT	ACGGGGGATG	5172
	GCGATAGACG	ACTTTGGTGA	CTTGGGCGAT	TCTGTGTGTC	GCAAATATCG	CAGTTTCGAT	5232
LO	ATAGGTGACA	GACGATATGA	GGCTATATCG	CCGATAGAGG	CGACATCAAG	CTGGCACATG	5292
	GCCAATGCAT	ATCGATCTAT	ACATTGAATC	AATATTGGCC	ATTAGCCATA	TTATTCATTG	5352
	GTTATATAGC	ATAAATCAAT	ATTGGCTATT	GGCCATTGCA	TACGTTGTAT	CCATATCATA	5412
	ATATGTACAT	TTATATTGGC	TCATGTCCAA	CATTACCGCC	ATGTTGACAT	TGATTATTGA	5472
· E	CTAGTTATTA	ATAGTAATCA	ATTACGGGGT	CATTAGTTCA	TAGCCCATAT	ATGGAGTTCC	5532
כו	GCGTTACATA	ACTTACGGTA	AATGGCCCGC	CTGGCTGACC	GCCCAACGAC	CCCCCCCAT	5592
	TGACGTCAAT	AATGACGTAT	GTTCCCATAG	TAACGCCAAT	AGGGACTTTC	CATTGACGTC	5652
	AATGGGTCCA	GTATTTACGG	TAAACTGCCC	ACTTGGCAGT	ACATCAAGTG	TATCATATGC	5712
	CAAGTACGCC	CCCTATTGAC	GTCAATGACG	GTAAATGGCC	CGCCTGGCAT	TATGCCCAGT	5772
20	ACATGACCTT	ATGGGACTTT	CCTACTTGGC	AGTACATCTA	CGTATTAGTC	ATCGCTATTA	5832
	CCATGGTGAT	GCGGTTTTGG	CAGTACATCA	ATGGGCGTGG	ATAGCGGTTT	GACTCACGGG	5892
	GATTTCCAAG	TCTCCACCCC	ATTGACGTCA	ATGGGAGTTT	GTTTTGGCAC	CAAAATCAAC	5952
	GGGACTTTCC	AAAATGTCGT	AACAACTCCG	CCCCATTGAC	GCAAATGGGC	GGTAGGCGTG	6012
25	TACGGTGGGA	GGTCTATATA	AGCAGAGCTC	GTTTAGTGAA	CCGTCAGATC	GCCTGGAGAC	6072
- /	GCCATCCACC	ChChhhhhCyC	СТССАТАСАА	GACACCCCCA	CCCATCCACC	CTCCCCCCC	6122

	GGGAACGGTG	CATTGGAACG	CGGATTCCCC	GTGCCAAGAG	TGACGTAAGT	ACCGCCTATA	6192
1	GAGTCTATAG	GCCCACCCC	TTGGCTTCTT	ATGCATGCTA	TACTGTTTTT	GGCTTGGGGT	62,52
	CTATACACCC	CCGCTTCCTC	ATGTTATAGG	TGATGGTATA	GCTTAGCCTA	TAGGTGTGGG	6312
	TTATTGACCA	TTATTGACCA	CTCCCCȚATT	GGTGACGATA	CTTTCCATTA	CTAATCCATA	6372
_	ACATGGCTCT	TTGCCACAAC	TCTCTTTATT	GGCTATATGC	CAATACACTG	TCCTTCAGAG	6432
5	ACTGACACGG	ACTCTGTATT	TTTACAGGAT	GGGGTCTCAT	TTATTATTTA	CAAATTCACA	6492
	TATACAACAC	CACCGTCCCC	AGTGCCCGCA	GTTTTTATTA	AACATAACGT	GGGATCTCCA	6552
	CGCGAATCTC	GGGTACGTGT	TCCGGACATG	GGCTCTTCTC	CGGTAGCGGC	GGAGCTTCTA	6612
	CATCCGAGCC	CTGCTCCCAT	CCCTCCAGCG	ACTCATGGTC	GCTCGGCAGC	TCCTTGCTCC	6672
10	TAACAGTGGA	GGCCAGACTT	AGGCACAGCA	CGATGCCCAC	CACCACCAGT	GTGCCGCACA	6732
	AGGCCGTGGC	GGTAGGGTAT	GTGTCTGAAA	ATGAGCTCGG	GGAGCGGGCT	TGCACCGCTG	6792
	ACGCATTIGG	AAGACTTAAG	GCAGCGGCAG	AAGAAGATGC	AGGCAGCTGA	GTTGTTGTGT	6852
	TCTGATAAGA	GTCAGAGGTA	ACTCCCGTTG	CGGTGCTGTT	AACGGTGGAG	GGCAGTGTAG	6912
1 =	TCTGAGCAGT	ACTCGTTGCT	GCCGCGCGCG	CCACCAGACA	TAATAGCTGA	CAGACTAACA	6972
15	GACTGTTCCT	TTCCATGGGT	CTTTTCTGCA	GTCACCGTCC	TTGACACGAA	GCTTGGGCTG	7032
	CAGGTCGATC	GACTCTAGAG	GATCGATCCC	CGGGCGAGCT	С		7073

### (2) INFORMATION FOR SEQ ID NO:16:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 219 amino acids
(B) TYPE: amino acid
(D) TOPOLOGY: linear 20

### (ii) MOLECULE TYPE: protein

25

### (xi) SEQUENCE DESCRIPTION: SEQ ID NO:16:

1 Gly Val His Ser Gln Val Gln Leu Val Glu Ser Gly Gly Val Val
1 5 10 15

Gln Pro Gly Arg Ser Leu Arg Leu Ser Cys Lys Ala Ser Gly Phe Agn

Gln Pro Gly Arg Ser Leu Arg Leu Ser Cys Lys Ala Ser Gly Phe Asn 20 25 30

Ile Lys Asp Tyr Tyr Met His Trp Val Arg Gln Ala Pro Gly Lys Gly
5 40 45

Leu Glu Trp Ile Gly Leu Ile Asp Pro Glu Asn Gly Asn Thr Ile Tyr 50 55 60

Asp Pro Lys Phe Gln Gly Arg Phe Ile Ile Ser Ala Asp Asn Ser Lys 65 70 75 80

Asn Thr Leu Phe Leu Gln Met Asp Ser Leu Arg Pro Glu Asp Thr Ala 10 85 90 95

Val Tyr Phe Cys Ala Arg Asp Asn Ser Tyr Tyr Phe Asp Tyr Trp Gly 100 105 110

Gln Gly Thr Pro Val Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser 115 120 125

Val Phe Pro Leu Ala Pro Cys Ser Arg Ser Thr Ser Glu Ser Thr Ala 135 130 140

Ala Leu Gly Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val 145 150 155 160

Ser Trp Asn Ser Gly Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala 165 170 175

Val Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val 20

Pro Ser Ser Ser Leu Gly Thr Lys Thr Tyr Thr Cys Asn Val Asp His 195 200 205

Lys Pro Ser Asn Thr Lys Val Asp Lys Arg Val 210 215

25

- (2) INFORMATION FOR SEQ ID NO:17:
- 1 (i) SEQUENCE CHARACTERISTICS:
  - (A) LENGTH: 12 amino acids
  - (B) TYPE: amino acid
  - (D) TOPOLOGY: linear
  - (ii) MOLECULE TYPE: protein
- 5 (xi) SEQUENCE DESCRIPTION: SEQ ID NO:17:

Glu Ser Lys Tyr Gly Pro Pro Cys Pro Ser Cys Pro 1 10

(2) INFORMATION FOR SEQ ID NO:18:

10

- (i) SEQUENCE CHARACTERISTICS:
  - (A) LENGTH: 109 amino acids
  - (B) TYPE: amino acid
  - (D) TOPOLOGY: linear
- (ii) MOLECULE TYPE: protein
- 15 (xi) SEQUENCE DESCRIPTION: SEQ ID NO:18:

Pro Glu Phe Leu Gly Gly Pro Ser Val Phe Leu Phe Pro Pro Lys Pro 1 10 15

Lys Asp Thr Leu Met Ile Ser Arg Thr Pro Glu Val Thr Cys Val Val 20 25 30

Val Asp Val Ser Gin Glu Asp Pro Glu Val Gin Phe Asn Trp Tyr Val 20 35 40 45

Asp Gly Val Glu Val His Asn Ala Lys Thr Lys Pro Arg Glu Glu Gln 50 55 60

Phe Asn Ser Thr Tyr Arg Val Val Ser Val Leu Thr Val Met His Gln 65 70 75 80

Asp Trp Leu Asn Gly Lys Glu Tyr Lys Cys Lys Val Ser Asn Lys Gly
25 90 95

Leu Pro Ser Ser Ile Glu Lys Thr Ile Ser Lys Ala Lys

1

5

- (2) INFORMATION FOR SEQ ID NO:19:
  - (i) SEQUENCE CHARACTERISTICS:
    - (A) LENGTH: 107 amino acids
    - (B) TYPE: amino acid
    - (D) TOPOLOGY: linear
  - (ii) MOLECULE TYPE: protein
  - (xi) SEQUENCE DESCRIPTION: SEQ ID NO:19:
- Gly Gln Pro Arg Glu Pro Gln Val Tyr Thr Leu Pro Pro Ser Gln Glu
  10 1 5 10 15

Glu Met Thr Lys Asn Gln Val Ser Leu Thr Cys Leu Val Lys Gly Phe 20 25 30

Tyr Pro Ser Asp Ile Ala Val Glu Trp Glu Ser Asn Gly Gln Pro Glu 35 45

Asn Asn Tyr Lys Thr Thr Pro Pro Val Leu Asp Ser Asp Gly Ser Phe 15 50 55 60

Phe Leu Tyr Ser Arg Leu Thr Val Asp Lys Ser Arg Trp Gln Glu Gly 65 70 75 80

Asn Val Phe Ser Val Ser Val Met His Glu Ala Leu His Asn His Tyr 85 90 95

Thr Gln Lys Ser Leu Ser Leu Ser Leu Gly Lys 20 100 105

- (2) INFORMATION FOR SEQ ID NO:20:
  - (i) SEQUENCE CHARACTERISTICS:
    - (A) LENGTH: 7864 base pairs
    - (B) TYPE: nucleic acid
- 25 (C) STRANDEDNESS: double
  - (D) TOPOLOGY: linear

(ii) MOLECULE TYPE: peptide

1

(ix) FEATURE:
(A) NAME/KEY: CDS
(B) LOCATION: 9..711

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:20:

5	AATTCACCAT	GGGTGTGCCA	ACTCAGGTAT	TAGGATTACT	GCTGCTGTGG	CTTACAGATG	60
	CAAGATGTGA	TATCCAAATG	ACACAATCTC	CTTCTTCTCT	AAGTGCTTCT	GTCGGAGATA	120
	GAGTAACAAT	TACATGTAAG	GCGAGTCAGG	ACATTAGAAA	GTATTTAAAC	TGGTATCAGC	180
	AAAAACCTGG	GAAGGCTCCT	AAGCTACTGA	TTTATTATGC	AACAAGTTTG	GCAGATGGAG	240
10	TACCTTCTAG	ATTTTCTGGT	TCTGGCTCTG	GAACAGACTA	CACATTCACA	ATTTCTTCTC	300
10	TCCAACCTGA	GGACATTGCT	ACATACTACT	GCCTACAACA	TGGTGAGAGT	CCGTATACAT	360
	TTGGACAAGG	AACAAAACTA	GAGATCACAA	GAACTGTTGC	GCCCCCTCT	GTCTTCATCT	420
	TCCCGCCATC	TGATGAGCAG	TTGAAATCTG	GAACTGCCTC	TGTTGTGTGC	CTGCTGAATA	480
	ACTTCTATCC	CAGAGAGGCC	AAAGTACAGT	GGAAGGTGGA	TAACGCCCTC	CAATCGGGTA	540
15	ACTCCCAGGA	GAGTGTCACA	GAGCAGGACA	GCAAGGACAG	CACCTACAGC	CTCAGCAGCA	600
	CCCTGACGCT	GAGCAAAGCA	GACTACGAGA	AACACAAAGT	CTACGCCTGC	GAAGTCACCC	660
	ATCAGGGCCT	GAGCTCGCCC	GTCACAAAGA	GCTTCAACAG	GGGAGAGTGT	TAGAGGGAGA	720
	AGTGCCCCCA	CCTGCTCCTC	AGTTCCAGCC	TGGGGATCAT	AATCAGCCAT	ACCACATTTG	780
20	TAGAGGTTTT	ACTTGCTTTA	AAAAACCTCC	CACACCTCCC	CCTGAACCTG	AAACATAAAA	840
20	TGAATGCAAT	TGTTGTTGTT	AACTTGTTTA	TTGCAGCTTA	TAATGGTTAC	AAATAAAGCA	900
	ATAGCATCAC	AAATTTCACA	AATAAAGCAT	TTTTTTCACT	GCATTCTAGT	TGTGGTTTGT	960
	CCAAACTCAT	CAATGTATCT	TATCATGTCT	GGATCCTCTA	CGCCGGACGC	ATCGTGGCCG	1020
	GCATCACCGG	CGCCACAGGT	GCGGTTGCTG	GCGCCTATAT	CGCCGACATC	ACCGATGGGG	1080
25	AAGATCGGGC	TCGCCACTTC	GGGCTCATGA	GCGCTTGTTT	CGGCGTGGGT	ATGGTGGCAG	1140

	GCCCGTGGCC	GGGGGACTGT	TGGGCGCCAT	CTCCTTGCAT	GCACCATTCC	TTGCGGCGGC	1200
1	GGTGCTCAAC	GGCCTCAACC	TACTACTGGG	CTGCTTCCTA	ATGCAGGAGT	CGCATAAGGG	1260
	AGAGCGTCGA	CCTCGGGCCG	CGTTGCTGGC	GTTTTTCCAT	AGGCTCCGCC	CCCTGACGA	1320
	GCATCACAAA	AATCGACGCT	CAAGTCAGAG	GTGGCGAAAC	CCGACAGGAC	TATAAAGATA	1380
5	CCAGGCGTTT	CCCCTGGAA	GCTCCCTCGT	GCGCTCTCCT	GTTCCGACCC	TGCCGCTTAC	1440
)	CGGATACCTG	TCCGCCTTTC	TCCCTTCGGG	AAGCGTGGCG	CTTTCTCAAT	GCTCACGCTG	1500
	TAGGTATCTC	AGTTCGGTGT	AGGTCGTTCG	CTCCAAGCTG	GGCTGTGTGC	ACGAACCCCC	1560
	CGTTCAGCCC	GACCGCTGCG	CCTTATCCGG	TAACTATCGT	CTTGAGTCCA	ACCCGGTAAG	1620
	ACACGACTTA	TCGCCACTGG	CAGCAGCCAC	TGGTAACAGG	ATTAGCAGAG	CGAGGTATGT	1680
10	AGGCGGTGCT	ACAGAGTTCT	TGAAGTGGTG	GCCTAACTAC	GGCTACACTA	GAAGGACAGT	1740
	ATTTGGTATC	TGCGCTCTGC	TGAAGCCAGT	TACCTTCGGA	AAAAGAGTTG	GTAGCTCTTG	1800
	ATCCGGCAAA	CAAACCACCG	CTGGTAGCGG	TGGTTTTTTT	GTTTGCAAGC	AGCAGATTAC	1860
	GCGCAGAAAA	AAAGGATCTC	AAGAAGATCC	TTTGATCTTT	TCTACGGGGT	CTGACGCTCA	1920
15	GTGGAACGAA	AACTCACGTT	AAGGGATTTT	GGTCATGAGA	TTATCAAAAA	GGATCTTCAC	1980
بــ	CTAGATCCTT	TTAAATTAAA	AATGAAGTTT	TAAATCAATC	TAAAGTATAT	ATGAGTAAAC	2040
	TTGGTCTGAC	AGTTACCAAT	GCTTAATCAG	TGAGGCACCT	ATCTCAGCGA	TCTGTCTATT	2100
	TCGTTCATCC	ATAGTTGCCT	GACTCCCCGT	CGTGTAGATA	ACTACGATAC	GGGAGGGCTT	2160
	ACCATCTGGC	CCCAGTGCTG	CAATGATACC	GCGAGACCCA	CGCTCACCGG	CTCCAGATTT	2220
20	ATCAGCAATA	AACCAGCCAG	CCGGAAGGGC	CGAGCGCAGA	AGTGGTCCTG	CAACTTTATC	2280
	CGCCTCCATC	CAGTCTATTA	ATTGTTGCCG	GGAAGCTAGA	GTAAGTAGTT	CGCCAGTTAA	2340
	TAGTTTGCGC	AACGTTGTTG	CCATTGCTAC	AGGCATCGTG	GTGTCACGCT	CGTCGTTTGG	2400
	TATGGCTTCA	TTCAGCTCCG	GTTCCCAACG	ATCAAGGCGA	GTTACATGAT	CCCCCATGTT	2460
25	GTGCAAAAAA	GCGGTTAGCT	CCTTCGGTCC	TCCGATCGTT	GTCAGAAGTA	AGTTGGCCGC	2520
- )	AGTGTTATCA	CTCATGGTTA	TGGCAGCACT	GCATAATTCT	CTTACTGTCA	TGCCATCCGT	2580

	AAGATGCTTT	TCTGTGACTG	GTGAGTACTC	AACCAAGTCA	TTCTGAGAAT	AGTGTATGCG	2640
1	GCGACCGAGT	TGCTCTTGCC	CGGCGTCAAC	ACGGGATAAT	ACCGCGCCAC	ATAGCAGAAC	2700
	TTTAAAAGTG	CTCATCATTG	GAAAACGTTC	TTCGGGGCGA	AAACTCTCAA	GGATCTTACC	2760
	GCTGTTGAGA	TCCAGTTCGA	TGTAACCCAC	TCGTGCACCC	AACTGATCTT	CAGCATCTTT	2820
5	TACTTTCACC	AGCGTTTCTG	GGTGAGCAAA	AACAGGAAGG	CAAAATGCCG	CAAAAAAGGG	2880
)	AATAAGGGCG	ACACGGAAAT	GTTGAATACT	CATACTCTTC	CTTTTTCAAT	ATTATTGAAG	2940
	CATTTATCAG	GGTTATTGTC	TCATGAGCGG	ATACATATTT	GAATGTATTT	AGAAAAATAA	3000
	ACAAATAGGG	GTTCCGCGCA	CATTTCCCCG	AAAAGTGCCA	CCTGACGTCT	AAGAAACCAT	3060
	TATTATCATG	ACATTAACCT	ATAAAAATAG	GCGTATCACG	AGGCCCTGAT	GGCTCTTTGC	3120
10	GGCACCCATC	GTTCGTAATG	TTCCGTGGCA	CCGAGGACAA	CCCTCAAGAG	AAAATGTAAT	3180
	CACACTGGCT	CACCTTCGGG	TGGGCCTTTC	TGCGTTTATA	AGGAGACACT	TTATGTTTAA	3240
	GAAGGTTGGT	AAATTCCTTG	CGGCTTTGGC	AGCCAAGCTA	GAGATCCGGC	TGTGGAATGT	3300
	GTGTCAGTTA	GGGTGTGGAA	AGTCCCCAGG	CTCCCCAGCA	GGCAGAAGTA	TGCAAAGCAT	3360
15	GCATCTCAAT	TAGTCAGCAA	CCAGGCTCCC	CAGCAGGCAG	aagtatgcaa	AGCATGCATC	3420
٠.	TCAATTAGTC	AGCAACCATA	GTCCCGCCCC	TAACTCCGCC	CATCCCGCCC	CTAACTCCGC	3480
	CCAGTTCCGC	CCATTCTCCG	CCCCATGGCT	GACTAATTTT	TTTTATTTAT	GCAGAGGCCG	3540
	AGGCCGCCTC	GGCCTCTGAG	CTATTCCAGA	AGTAGTGAGG	AGGCTTTTTT	GGAGGCCTAG	3600
	GCTTTTGCAA	AAAGCTAGCT	TGGGGCCACC	GCTCAGAGCA	CCTTCCACCA	TGGCCACCTC	3660
20	AGCAAGTTCC	CACTTGAACA	AAAACATCAA	GCAAATGTAC	TTGTGCCTGC	CCCAGGGTGA	3720
	GAAAGTCCAA	GCCATGTATA	TCTGGGTTGA	TGGTACTGGA	GAAGGACTGC	GCTGCAAAAC	3780
	CCGCACCCTG	GACTGTGAGC	CCAAGTGTGT	AGAAGAGTTA	CCTGAGTGGA	ATTTTGATGG	3840
	CTCTAGTACC)	TTTCAGTCTG	AGGGCTCCAA	CAGTGACATG	TATCTCAGCC	CTGTTGCCAT	3900
25 .	GTTTCGGGAC	CCCTTCCGCA	GAGATCCCAA	CAAGCTGGTG	TTCTGTGAAG	TTTTCAAGTA	3960
. ر-	CAACCGGAAG	CCTGCAGAGA	CCAATTTAAG	GCACTCGTGT	AAACGGATAA	TGGACATGGT	4020

	GAGCAACCAG	CACCCCTGGT	TTGGAATGGA	ACAGGAGTAT	ACTCTGATGG	GAACAGATGG	4080
1	GCACCCTTTT	GGTTGGCCTT	CCAATGGCTT	TCCTGGGCCC	CAAGGTCCGT	ATTACTGTGG	4140
	TGTGGGCGCA	GACAAAGCCT	ATGGCAGGGA	TATCGTGGAG	GCTCACTACC	GCGCCTGCTT	4200
	GTATGCTGGG	GTCAAGATTA	CAGGAACAAA	TGCTGAGGTC	ATGCCTGCCC	AGTGGGAACT	4260
5	CCAAATAGGA	CCCTGTGAAG	GAATCCGCAT	GGGAGATCAT	CTCTGGGTGG	CCCGTTTCAT	4320
כ	CTTNCATCGA	GTATGTGAAG	ACTTTGGGGT	AATAGCAACC	TTTGACCCCA	AGCCCATTCC	4380
	TGGGAACTGG	AATGGTGCAG	GCTGCCATAC	CAACTTTAGC	ACCAAGGCCA	TGCGGGAGGA	4440
	GAATGGTCTG	AAGCACATCG	AGGAGGCCAT	CGAGAAACTA	AGCAAGCGGC	ACCGGTACCA	4500
	CATTCGAGCC	TACGATCCCA	AGGGGGGCCT	GGACAATGCC	CGTGGTCTGA	CTGGGTTCCA	4560
10	CGAAACGTCC	AACATCAACG	ACTITICTGC	TGGTGTCGCC	AATCGCAGTG	CCAGCATCCG	4620
	CATTCCCCCG	ACTGTCGGCC	AGGAGAAGAA	AGGTTACTTT	GAAGACCGCG	GCCCTCTGC	4680
	CAATTGTGAC	CCCTTTGCAG	TGACAGAAGC	CATCGTCCGC	ACATGCCTTC	TCAATGAGAC	4740
	TGGCCACGAG	CCCTTCCAAT	ACAAAAACTA	ATTAGACTTT	GAGTGATCTT	GAGCCTTTCC	4800
15	TAGTTCATCC	CACCCCCCCC	CAGAGAGATC	TTTGTGAAGG	AACCTTACTT	CTGTGGTGTG	4860
עב	ACATAATTGG	ACAAACTACC	TACAGAGATT	TAAAGCTCTA	AGGTAAATAT	ATTTTTA	4920
	AGTGTATAAT	GTGTTAAACT	ACTGATTCTA	ATTGTTTGTG	TATTTTAGAT	TCCAACCTAT	4980
	GGAACTGATG	AATGGGAGCA	GTGGTGGAAT	GCCTTTAATG	AGGAAAACCT	GTTTTGCTCA	5040
	GAAGAAATGC	CATCTAGTGA	TGATGAGGCT	ACTGCTGACT	CTCAACATTC	TACTCCTCCA	5100
20	AAAAAGAAGA	GAAAGGTAGA	ACACCCCAAG	GACTTTCCTT	CAGAATTGCT	AAGTTTTTTG	5160
	AGTCATGCTG	TGTTTAGTAA	TAGAACTCTT	GCTTGCTTTG	CTATTTACAC	CACAAAGGAA	5220
	AAAGCTGCAC	TGCTATACAA	GAAAATTATG	GAAAAATATT	CTGTAACCTT	TATAAGTAGG	5280
		•	CATACTGTTT			·	5340
25	•		AAAATTGTGT				5400
_	AATAAGGAAT	ATTTGATGTA	TAGTGCCTAG	ACTAGAGATC	ATAATCAGCC	ATACCACATT	5460

	TGTAGAGGTT	TTACTTCCTT	TAAAAAACCT	CCCACACCTC	CCCCTGAACC	TGAAACATAA	5520
1	AATGAATGCA	ATTGTTGTTG	TTAACTTGTT	TATTGCAGCT	TATAATGGTT	ACAAATAAAG	5580
	CAATAGCATC	ACAAATTTCA	CAAATAAAGC	ATTTTTTCA	CTGCATTCTA	GTTGTGGTTT	5640
	GTCCAAACTC	ATCAATGTAT	CTTATCATGT	CTGGATCTCT	AGCTTCGTGT	CAAGGACGGT	5700
5	GACTGCAGTG	AATAATAAA	TGTGTGTTTG	TCCGAAATAC	GCGTTTTGAG	ATTTCTGTCG	5760
)	CCTACTAAAT	TCATGTCGCG	CGATAGTGGT	GTTTATCGCC	GATAGAGATG	GCGATATTGG	5820
	AAAAATCGAT	ATTTGAAAAT	ATGGCATATT	GAAAATGTCG	CCGATGTGAG	TTTCTGTGTA	5880
	ACTGATATCG	CCATTTTTCC	AAAAGTGATT	TTTGGGCATA	CGCGATATCT	GGCGATAGCG	5940
	CTTATATCGT	TTACGGGGGA	TGGCGATAGA	CGACTTTGGT	GACTTGGGCG	ATTCTGTGTG	6000
10	TCGCAAATAT	CGCAGTTTCG	ATATAGGTGA	CAGACGATAT	GAGGCTATAT	CGCCGATAGA	6060
	GGCGACATCA	AGCTGGCACA	TGGCCAATGC	ATATCGATCT	ATACATTGAA	TCAATATTGG	6120
	CCATTAGCCA	TATTATTCAT	TGGTTATATA	GCATAAATCA	ATATTGGCTA	TTGGCCATTG	6180
	CATACGTTGT	ATCCATATCA	TAATATGTAC	ATTTATATTG	GCTCATGTCC	AACATTACCG	6240
15	CCATGTTGAC	ATTGATTATT	GACTAGTTAT	TAATAGTAAT	CAATTACGGG	GTCATTAGTT	·6300
- /	CATAGCCCAT	ATATGGAGTT	CCGCGTTACA	TAACTTACGG	TAAATGGCCC	GCCTGGCTGA	6360
	CCGCCCAACG	ACCCCCCCCC	ATTGACGTCA	ATAATGACGT	ATGTTCCCAT	AGTAACGCCA	6420
	ATAGGGACTT	TCCATTGACG	TCAATGGGTG	GAGTATTTAC	GGTAAACTGC	CCACTTGGCA	6480
	GTACATCAAG	TGTATCATAT	GCCAAGTACG	CCCCCTATTG	ACGTCAATGA	CGGTAAATGG	6540
20 ·	, CCCGCCTGGC	ATTATGCCCA	GTACATGACC	TTATGGGACT	TTCCTACTTG	GCAGTACATC	6600
	TACGTATTAG	TCATCGCTAT	TACCATGGTG	ATGCGGTTTT	GGCAGTACAT	CAATGGGCGT	6660
	GGATAGCGGT	TTGACTCACG	GGGATTTCCA	AGTCTCCACC	CCATTGACGT	CAATGGGAGT	6720
	TTGTTTTGGC	ACCAAAATCA	ACGGGACTTT	CCAAAATGTC	GTAACAACTC	CGCCCCATTG	6780
25	ACGCAAATGG	GCGGTAGGCG	TGTACGGTGG	GAGGTCTATA	TAAGCAGAGC	TCGTTTAGTG	6840
_	AACCCTCACA	TOCCOTOCAC	ACCCCATCCA	CCCatiCataladatic	ACCTCCATAG	AAGACACCCC	6900

	GACCGATCCA	GCCTCCGCGG	CCGGGAACGG	TGCATTGGAA	CGCGGATTCC	CCGTGCCAAG	6960
1	AGTGACGTAA	GTACCGCCTA	TAGAGTCTAT	AGGCCCACCC	CCTTGGCTTC	TTATGCATGC	7020
	TATACTGTTT	TTGGCTTCGG	GTCTATACAC	CCCCGCTTCC	TCATGTTATA	GGTGATGGTA	7080
	TAGCTTAGCC	TATAGGTGTG	GGTTATTGAC	CATTATTGAC	CACTCCCCTA	TTGGTGACGA	7140
5	TACTTTCCAT	TACTAATCCA	TAACATGGCT	CTTTGCCACA	ACTCTCTTTA	TTGGCTATAT	7200
	GCCAATACAC	TGTCCTTCAG	AGACTGACAC	GGACTCTGTA	TTTTTACAGG	ATGGGGTCTC	7260
	ATTTATTATT	TACAAATTCA	CATATACAAC	ACCACCGTCC	CCAGTGCCCG	CAGTTTTTAT	7320
	TAAACATAAC	GTGGGATCTC	CACGCGAATC	TCGGGTACGT	GTTCCGGACA	TGGGCTCTTC	7380
	TCCGGTAGCG	GCGGAGCTTC	TACATCCGAG	CCCTGCTCCC	ATGCCTCCAG	CGACTCATGG	7440
10	TCGCTCGGCA	TCTCCTTGCT	CCTAACAGTG	GAGGCCAGAC	TTAGGCACAG	CACGATGCCC	7500
	ACCACCACCA	GTGTGCCGCA	CAAGGCCGTG	GCGGTAGGGT	ATGTGTCTGA	AAATGAGCTC	7560
	GGGGAGCGGG	CTTGCACCGC	TGACGCATTT	GGAAGACTTA	AGGCAGCGGC	AGAAGAAGAT	7620
	GCAGGCAGCT	GAGTTGTTGT	GTTCTGATAA	GAGTCAGAGG	TAACTCCCGT	TGCGGTGCTG	7680
15	TTAACGGTGG	AGGGCAGTGT	AGTCTGAGCA	GTACTCGTTG	CTGCCGCGCG	CGCCACCAGA	7740
-,	CATAATAGCT	GACAGACTAA	CAGACTGTTC	CTTTCCATGG	GTCTTTTCTG	CAGTCACCGT	7800
	CCTTGACACG	AAGCTTGGGC	TGCAGGTCGA	TCGACTCTAG	AGGATCGATC	CCCGGGCGAG	7860
	CTCG						7864

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### WHAT IS CLAIMED IS:

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- A CDR-grafted antibody capable of inhibiting human tissue factor wherein the complementarity determining regions (CDRs) are derived from a non-human monoclonal antibody against tissue factor and the framework (FR) and constant (C) regions are derived from one or more human antibodies.
- 2. The CDR-grafted antibody of Claim 1 wherein said non-human monoclonal antibody is a murine 10 antibody.
  - 3. The CDR-grafted antibody of Claim 2 wherein said murine antibody is TF8-5G9.
- 4. The CDR-grafted antibody of Claim 1 wherein said CDRs of the heavy chain have the amino acid 15 sequences:

CDR1 DDYMH (SEQ ID NO:5)

CDR2 LIDPENGNTIYDPKFQG (SEQ ID NO:6)

CDR3 DNSYYFDY (SEQ ID NO:7)

and said CDRs of the light chain have the amino acid

20 sequences:

CDR1		KASQDIRKYLN	•	(SEQ	ID	NO:8.)
CDR2		YATSLAD		-(SEQ	ID	NO:9)
CDR3	•	LQHGESPYT		(SEO	ID	NO:10).

- 5. The CDR-grafted antibody of Claim 1
  25 wherein the FR of the heavy chain is derived from the human antibody KOL.
  - 6. The CDR-grafted antibody of Claim 1 wherein the FR of the light chain is derived from the human antibody REI.

- 7. The CDR-grafted antibody of Claim 1 1 wherein the heavy chain variable region has the amino acid sequence of SEQ ID NO:11.
- 8. The CDR-grafted antibody of Claim 1 or 7 wherein the light chain variable region has the amino 5 acid sequence of SEQ ID NO:12.
  - 9. The CDR-grafted antibody of Claim 1 wherein the heavy chain variable region has the amino acid sequence of SEQ ID NO:13.
- 10. The CDR-grafted antibody of Claim 1 or 9 10 wherein the light chain variable region has the amino acid sequence of SEQ ID NO:14.
  - 11. The CDR-grafted antibody of Claim 1 wherein the heavy chain constant region is the human IgG4 constant region.
- 15 12. The CDR-grafted antibody of Claim 10 wherein the heavy chain constant region is the human IgG4 constant region.
- 13. The CDR-grafted antibody of Claim 1 wherein the light chain constant region is the human 20 kappa constant region.
  - 14. The CDR-grafted antibody of Claim 10 wherein the light chain constant region is the human kappa constant region.
- 15. CDR-grafted monoclonal antibody TF8HCDR1 25 x TF8LCDR1.
  - 16. CDR-grafted monoclonal antibody TF8HCDR20 x TF8LCDR3.
- 17. A fragment of the CDR-grafted antibody of Claim 1 wherein said fragment is capable of inhibiting 30 human tissue factor.

- 18. The fragment of Claim 17 wherein said 1 fragment is an Fab or F(ab'), fragment.
- antibody of Claim 1 comprising cotransfecting a host cell with an expression vector comprising a nucleic acid encoding the CDR-grafted antibody heavy chain and an expression vector comprising a nucleic acid encoding the CDR-grafted antibody light chain; culturing the transfected host cell; and recovering said CDR-grafted antibody.
- 20. A method of making the CDR-grafted antibody of Claim 1 comprising transfecting a host cell with an expression vector comprising a nucleic acid encoding the CDR-grafted antibody heavy chain and a nucleic acid encoding the CDR-grafted antibody light chain; culturing the transfected host cell; and recovering said CDR-grafted antibody.
- 21. The method of Claim 18 or 19 wherein said nucleic acid encoding the CDR-grafted antibody heavy chain has the sequence of nucleotides 1-2360 of SEQ ID 20 NO:15.
  - 22. The method of Claim 18 or 19 wherein said nucleic acid encoding the CDR-grafted light chain has the sequence of nucleotides 1-759 of SEQ ID NO:17.
- 23. The method of Claim 19 or 20 wherein said 25 host cell is a bacterial cell, yeast cell, insect cell or mammalian cell.
  - 24. The method of Claim 23 wherein said mammalian cell is a CHO cell, COS cell or myeloma cell.
- 25. The method of Claim 19 wherein said 30 expression vector comprising a nucleic acid encoding the CDR-grafted antibody heavy chain is pEe6TF8HCDR20.

- 26. The method of Claim 19 wherein said l expression vector comprising a nucleic acid encoding the CDR-grafted antibody light chain is pEel2TF8LCDR3.
  - 27. A nucleic acid encoding the heavy chain of the CDR-grafted antibody of Claim 1.
- 5 28. A nucleic acid encoding the light chain of the CDR-grafted antibody of Claim 1.
  - 29. The nucleic acid of Claim 27 having the sequence of nucleotides 1-2360 of SEQ ID NO:15.
- 30. The nucleic acid of Claim 28 having the 10 sequence of nucleotides 1-759 of SEQ ID NO:17.
- 31. A method of attenuation of coagulation comprising administering a therapeutically effective amount of a CDR-grafted antibody capable of inhibiting human tissue factor to a patient in need of said attenuation.
  - 32. The method of Claim 31 wherein said CDRgrafted antibody is TF8HCDR20 x TF84CDR3.
- 33. A method of treatment or prevention of thrombotic disorder comprising administering a
  20 therapeutically effective amount of a CDR-grafted antibody capable of inhibiting human tissue factor to a patient in need of said treatment or prevention.
- 34. The method of Claim 33 wherein said thrombotic disorder is intravascular coagulation,25 arterial restenosis or arteriosclerosis.
  - 35. The method of Claim 33 or 34 wherein said CDR-grafted antibody is TF8HCDR20 x TF8LCDR3.
- 36. A pharmaceutical composition comprising at least one CDR-grafted antibody capable of inhibiting 30 human tissue factor and a pharmaceutically acceptable carrier.

37. The pharmaceutical composition of Claim 1 36 wherein said CDR-grafted antibody is TF8HCDR20  $\times$  TF8LCDR3.

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1/41

Sequence of the murine TF8-5G9 heavy chain cDNA with protein translation. The essential regions of the cDNA are as follows:

•	<u>Nucleotides</u>	Region
EIC 1 A	1-10	5' untranslated region.
FIG. 1 A	11-67	Start codon and leader sequence.
	68-418	Variable region.
	419-1390	Murine IgG1 constant region.
	1391-1489	3' untranslated region.

Sequence Range: 1 to 1489

				10			20			3	0			40			
				•			•				• 			•			
	CCI	CCT	TAC	A A:	IG A	M T	GC AI	GC IN	GC G	TC X	IC T	IC T	IC C	LC Y.	TC C	CA GTO	;
		GLAR	ATG	TT	AC T	rr A	CC T			AG T	NG A	AG A	AG G	AC T	YC C	CA GIT	:
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	Val	Thr	Gly	Val	λsn	Ser	Glu	Ile	Gln	Leu	Gln	Gln	Ser	Glv	λla	Glu>	
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	TTC	YYC	λTT	λλλ	CAC	TAC	TAT	λTG	CAC	TGG	GTG	AAG	CAG	100	-		
	~~	116	TAA	TIT	CIG	ATG	ATA	TAC	CTY:	ACC	CAC	مكلمك		TO 0			
	Phe	Asn	Ile	Lys	Yab	Tyr	Tyr	Met	His	Trp	Val	Lys	Gln	λεσ	Pro	Glu>	
		20	30		•	210			220			2:	30		:	240	
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	GTC	CCC	GAC		) CC	TAL	CCL	116	ATT	GAT	CCT	CXC	AAT	CCT	λλT	ACT	
	Gln	Gly	Leu	Glu	Tro	Ile	Glv	Leu	TIA	CIN	D	CIC	TTA	CCY	TTA	TGA Thr>	
							,		110	nop	FIO	GIR	ABI	GIA	Yed	Thr>	
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	IVI	AIA	CIC	CCC	TIC	XXC	crc	CCC	TTC	CCG	TCA	TAT	TYPE	C T	<b>CTT</b>	T-T	
	Ile	Tyr	Asp	Pro	Lys	Phe	Gln	Gly	Lys	λla	Ser	Ile	Thr	λla	ARD	Thr>	
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	<b>TYY</b>	<b>TCC</b>	110	101	-~		-				•			•			
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	Ser	AGG Ser	Age	Thr	216	TUE	LAC	GIC	UAG Tarr	TUI Par	TCC	GAC	TCT	yCY	CIC	CIG	
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## FIG. 1 B

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	TCC	_	CAA	GGC	» »CC	3 Cm		1 C 1	•	<b>5</b> 00		•				
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	Trp	Gly	Gln	Gly	Thr	Thr	Léu	Thr	Val	Ser	Ser	λla	Lys	Thr	Thr	GGG Pro>
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	CCA	TCT	GTC	TAT	CCA	CIK	CCC	بلمان	CCA	יואיינו	CCAL		•			•
	CCT		فللاث	ATA	CGT	GAC	CCC	GGA	CCT	XCX	CC3	CCC		TV-13	_	
	Pro	Ser	Val	Tyr	Pro	Leu	λla	Pro	Gly	Ser	Ala	λla	Cln	Thr	λen	AGG Ser>
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			490			50	•			10			520			
	λTG	GIG	ACC	CIC	CCA	TGC	CIC	CIC	λλG	CCC	TAT	TTC	CCI	CAG	CCX	GTG
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	<b>ACA</b>	CTC	ACC	TCC	λλС	TCT	GGA	TCC	CTG	TCC	AGC	CCT	crc	CAC	ACC	TTC
	TCT	CXC	TCC	YCC	TTG	YCY	CCT	YCC	CYC	YCC	TCC	CCY	CAC	GTG	TGG	AAG
•	THE	AST	Thr	TIP	ABI	Ser	GIA	Ser	ren	Ser	Ser	CIA	Val	His	Thr	Phe>
	580			59	0		6	500			610			62	20	
	CCA	ccr	CTC	CIG.	• CXG	TCT	CYC	CIC	TAC	ACT	crc	λGC	AGC	TCA	GTG	ACT
	CCA CCT	CCY	CXC	CTG GAC	cxc	λGλ	GAC	CIC	ATG	TCA	CTG GAC	TCG	TCG	TCA AGT	GTG CAC	TGA
	CCA CCT	CCY	CXC	CTG GAC	cxc	λGλ	GAC	CIC	ATG	TCA	CTG GAC	TCG	TCG	TCA AGT	GTG CAC	ACT TGA Thr>
	CCA GGT Pro	CCY	CXC	CTG GAC	cxc	λGλ	GAC	CTC GAG Leu	ATG	TCA	CTG GAC Lou	TCG	TCG	TCA AGT	GTG CAC	TGA
	CCA GGT Pro	CCC CCC	TCC Val	CTG GAC Leu	CAG GTC Glm 640	AGA Ser TGG	CCC	CTC GAG Leu 6!	ATG Tyr 50 CAG	TOA	CTG GAC Lett	TCG Ser 660	TCC Ser TCC	TCA AGT Ser	GTG CAC Val 670	TGA Thr>
	CCA CGT PTO	CCA Ala 630 CCC CCC	TCC AGG	CTG GAC Leu AGC TCG	CAG GTC Glm 640 ACC	AGA Ser TGG ACC	GAC CTG AMP CCC CCC	CTC GAG Leu 6! AGC TCG	ATG Tyr 50 CAG CTC	TGA Thr ACC TGG	CTG GAC Loni	TCG Ser 660 ACC TCG	TCC Ser TCC ACC	TCA AGT Ser	GTG CAC Val 670 GTT CAA	TGA Thr>
	CCA CGT PTO	CCA Ala 630 CCC CCC	TCC AGG	CTG GAC Leu AGC TCG	CAG GTC Glm 640 ACC	AGA Ser TGG ACC	GAC CTG AMP CCC CCC	CTC GAG Leu 6! AGC TCG	ATG Tyr 50 CAG CTC	TGA Thr ACC TGG	CTG GAC Loni	TCG Ser 660 ACC TCG	TCC Ser TCC ACC	TCA AGT Ser	GTG CAC Val 670 GTT CAA	TGA Thr>
	CCA CGT PTO	CGA Ala 630 CCC GGG PEO	TCC AGG	CTG GAC Leu AGC TCG	CAG GTC Gln 640 ACC TGG	AGA Ser TGG ACC	GAC CTG AMP CCC CCC	CTC GAG Leu 6! AGC TCG	ATG Tyr 50 CAG CTC	TGA Thr ACC TGG	CTG GAC Loni	TCG Ser 660 ACC TGG Thr	TCC Ser TCC ACC	TCA AGT Ser	GTG CAC Val 670 GTT CAA Val	TGA Thr>
	CCA CGT PTO CTC CAC Val	CCA Ala 630 CCC GGG PEO 60	TCC AGG Ser	CTG GAC Leu AGC TCG Ser	CAG GTC Gln 640 ACC TGG Thr	TCG ACC TIP	CAC CTG Amp CCC GGG PTO	CTC GAG Leu 69 AGC TCG Ser	ATG Tyr 50 CAG CTC Glu 700	TGA Thr ACC TGG Thr	CTG GAC Long	TCG Ser 660 ACC TCG Thr 7:	TCC Ser TCC ACC Cys	TCA AGT Ser AAC TTG ABD	GTG CAC Val 670 GTT CAA Val	GCC CCG Ala>
	CCA CGT PTO CAC CAC Val	CCA Ala 630 CCC GGG PEO 60 CCG GGG CCG CCG	TCC AGG Ser GCC CGG	AGC TCG	CAG GTC Gln 640 ACC TGG Thr	TGG ACC TTP	GAC CTG AMP CCC GGG PTO	CTC GAG Leu 6! ACC TCG Ser GTC CAC	ATG TYE  GAG CTC Glu  700 GAC CTG	TGA Thr ACC TGG Thr AAG TTC	CTG GAC Loni CTC CAG Val	TCG Ser 660 ACC TCG Thr 7:	TCC Ser TCC ACG CyB	TCA AGT Ser AAC TTG ABD	GTG CAC Val 670 GTT CAA Val	TGA Thr> GCC CGG Ala> 720 GAT CTA
	CCA CGT PTO CAC CAC Val	CCA Ala 630 CCC GGG PEO 60 CCG GGG CCG CCG	TCC AGG Ser GCC CGG	AGC TCG	CAG GTC Gln 640 ACC TGG Thr	TGG ACC TTP	GAC CTG AMP CCC GGG PTO	CTC GAG Leu 6! ACC TCG Ser GTC CAC	ATG Tyr 50 CAG CTC Glu 700 GAC CTG	TGA Thr ACC TGG Thr AAG TTC	CTG GAC Loni CTC CAG Val	TCG Ser 660 ACC TCG Thr 7:	TCC Ser TCC ACG CyB	TCA AGT Ser AAC TTG ABD	GTG CAC Val 670 GTT CAA Val	GCC CCG Ala>
	CCA CGT PTO CAC CAC Val	CCA Ala 630 CCC GGG PEO 60 CCG GGG CCG CCG	TCC AGG Ser GCC CGG	AGC TCG Ser	CAG GTC Gln 640 ACC TGG Thr	TGG ACC TIP ACC TGG Thr	GAC CTG AMP CCC GGG PTO	CTC GAG Leu 6! ACC TCG Ser GTC CAC	ATG TYE  GAG CTC Glu  700 GAC CTG ABP	TGA Thr ACC TGG Thr AAG TTC	CTG GAC Loni CTC CAG Val	TCG Ser 660 ACC TCG Thr 7:	TCC Ser TCC ACG CyB	AAC TIG ASD	GTG CAC Val 670 GTT CAA Val	TGA Thr> GCC CGG Ala> 720 GAT CTA
	CCA CGT Pro CAC CAC Val	CCA Ala 630 CCC GGG PTO 66 CCG GGC PTO	TCC AGG Ser 80 . GCC CGG Ala 730 . TGT	AGC TCG Ser	CAG GTC Glm 640 ACC TGG Thr AGC TCG Ser	AGA Ser TGG ACC TTP 690 ACC TGG Thr	CAC CTG AMP CCC GGG PTO AAG TTC Lys	CTC GAG Letu 65 AGC TCG Ser GTC CAC CAC CAC TCT CTC CTC CTC CTC CTC C	ATG Tyr GAG CTC Glu 700 GAC CTG ABP	TGA Thr ACC TGG Thr AAG TTC Lys	CTC CAG Lots Val	TCG Ser 660 TCG TCG Thr 7: ATT TAA Ile	TCG Ser TGC ACG CyB 10 GTG CAC Val 760	TCA AGT Ser AAC TTG Asn CCC GGG PTO	GTG CAC Val 670 CAA Val AGG TCC ATT	GCC CGG Ala> 720 GAT CTA ABP>
	CCA CGT PTO CAC CAC CTG His	CCA Ala 630 CCC GGG PTD 60 CCG GGC PTD	TCC AGG Ser 80 . GCC CGG Ala 730 . TGT ACA	AGC TCG Ser	CAG GTC Glm 640 ACC TCG TCG TCG Ser	AGA Ser TGG ACC TTP 690 ACC TGG Thr	CAC CTG AMP CCC GGG PTO AAG TTC Lym	CTC GAG Leu 6: AGC TCG Ser GTC CAC Val	ATG Tyr GAG CTC Glu 700 GAC CTG ABP	TGA Thr ACC TGG Thr AAG TTC Lyss 750 GTC CAG	CTC GAC Loui	TCG Ser 660 TCG Thr 7: ATT TAA Ile	TCC Ser TCC ACC Cys CAC Val 760 CAT CAT	TCA AGT Ser AAC TTG ABD CCC GGG PTO	GTG CAC Val 6700 CAA Val AGG TCC ATY	TCA Thr>  GCC CGG Ala>  720 GAT CTA Asp> GTC CAG
	CCA CGT PTO CAC CAC CTG His	CCA Ala 630 CCC GGG PTD 60 CCG GGC PTD	TCC AGG Ser 80 . GCC CGG Ala 730 . TGT ACA	AGC TCG Ser	CAG GTC Glm 640 ACC TCG TCG TCG Ser	AGA Ser TGG ACC TTP 690 ACC TGG Thr	CAC CTG AMP CCC GGG PTO AAG TTC Lym	CTC GAG Leu 6: AGC TCG Ser GTC CAC Val	ATG Tyr GAG CTC Glu 700 GAC CTG ABP	TGA Thr ACC TGG Thr AAG TTC Lyss 750 GTC CAG	CTC GAC Loui	TCG Ser 660 TCG Thr 7: ATT TAA Ile	TCC Ser TCC ACC Cys CAC Val 760 CAT CAT	TCA AGT Ser AAC TTG ABD CCC GGG PTO	GTG CAC Val 6700 CAA Val AGG TCC ATY	GCC CGG Ala> 720 GAT CTA ABP>
7	CCA CGT PTO CAC CAC CTG His	CCA Ala 630 CCC GGG PTD 60 CCG GGC PTD	TCC AGG Ser GCC CGG Ala 730 TGT ACA CYB	AGC TCG Ser	CAG GTC Glm 640 ACC TCG TCG TCG Ser	AGA Ser TGG ACC TTP 690 ACC TGG Thr	CAC CTG AMP CCC GGG PTO AAG TTC Lym	CTC GAG Leu 6: AGC TCG Ser GTC CAC Val	ATG Tyr GAG CTC Glu 700 GAC CTG ABP	ACC TICK TICK LYB	CTC GAC Loui	TCG Ser 660 TCG Thr 7: ATT TAA Ile	TCC Ser TCC ACC Cys CTC CAC CYs T60 CTC CAC CAT Val	TCA AGT Ser AAC TTG ABD CCC GGG PTO	GTG CAC Val 6700 CAA Val AGG TCC ATY	TCA Thr>  GCC CGG Ala>  720 GAT CTA Asp> GTC CAG
7.	CCA CAC CAC CAC CAC TOTAL CAC CAC TTC TTC	CCA Ala 630 CCC GGG PTO 60 CCG GGC PTO CCA Gly ATC	TCC AGG Ser GCC CGG Ala 730 . TCT ACA Cys	AGC TCG Ser  AGC TCG Ser  AGC TCG Ser  AGC TCG Ser  AGC TCG Ser	CAG GTC Gln 640 ACC TCG TCG TCG Ser CCT GGA PTD	AGA Ser TGG ACC TTP 690 ACC TGG Thr TGG ACG Cys	CAC CTG AMP CCC GGG PTO AAG TTC Lym 40 ATA TAT Ile 790	CTC GAG Lett 6: AGC TCG CAC Val	ATG TYT  GAG CCTC GGlu  7000 GAC CTG ASP  ACA TGT Thr	TGA Thr ACC TCG TCG Thr AAG TTC. Lys  CTC CAG Val  STC	CTC GAC Loui GAC CAG CAG CAG CAG CAG CAG CAG CAG CAG	TCG Ser 660 ACC TCG Thr 77 ATT TAA Ile	TCC Ser TCC ACC Cys 10 CTC CAC Val CTAT Val	ACT Ser AAC TTC AAST ACT Ser ACT	GTG CAC Val 670 CAA Val AGG TCC AITY TCT AGA Ser	CCC CCG Ala> 720 CAT CTA Asp> CTC CAG Val>
7	CCA COTC PTO CAC CAC CAC CAC CAC CAC CAC CAC CAC CA	CCA Ala 630 CCC GGG PTO 60 CCG GGC PTO CCA Gly ATC TAG	TCC AGG Ser GCC CGG Ala 730 . TCT ACA Cys	AGC TCG Ser  AGC TCG Ser  AGC TCG Ser  AGC TCG Ser  CCC CGC	CAG GTC Gln 640 ACC TCG TCG TCG Ser CCT GGA PTD	AGA Ser TGG ACC TTP 690 ACC TGG TCG Cys	CAC CTG AMP  CCC GGG Pro  AAG TTC Lym  ATA TAT Ile  790  CCC GGG	CTC GAG Lett 6: AGC TCG CAC Val TGT ACA CYS	ATG TYT  GAG CCTC GGlu  7000 GAC CTG AEP  ACA TGT Thr  CAT CTA	ACC TOC TOC Lys	CTC CAG	TCG Ser 660 ACC TCG Thr 77 ATT TAA Ile GAA CTT Glu	TCC Ser  TCC ACC Cys  CTC CAC Val  T60 CAT Val  ATT TAA	ACC CCC CCC CCC PIO	GTG CAC Val 670 CAA Val AGG TCC ATT AGA Ser	TCA Thr>  GCC CGG Ala>  720 GAT CTA Asp> GTC CAG

## RECTIFIED SHEET (RULE 91)

ISA/EP

## FIG. 1 C

1	820			a	30		1	840			850		-	8	60	
1	œ	AAG	GTC	λŒ	TCT	CIT	CIG	GTA	CYC	ATC	AGC	λλG	CAT	CAT	œ	CAG
1	CCA Pro	ITC	Val	TGC	YCY	CAA	CAC	CAT	CIG	TAG	For	IV	CIA	CIA	CCC	CIC CIC
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		870			880			. 8	90.			900			910	
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				261	H	Phe	Val	AND	ABD	AST	Glu	Val	His	Thr	λla	GIC .
		-	20			930			940				50			960
	λŒ	CAA	ccc	CCC	GAG	GAG	CAG	TTC	λλC	λGC	ACT	TTC	œc	TCA	GTC	λGT
	100	611		تتاعا	CTT.	(714.	CTIC	AAC	تكلمك		T-2	330				
		`.			GIU	GIU	GIII	1110	ABII	SEL	Thr	Phe	yrg	Ser	Val	TCA Ser>
			970 *		•	_	80			990			1000			
	CAN	CIT	CCC	ATC	ATG	CAC	CYC	CAC	TGG	CIC	AAT	CCC	λλG	CXC	TTC	<b>XXX</b> .
	Glu	Lou	Pro	Ile	Met	His	Gln	Asp	TIP	Lou	TTA Am	CCC	TTC	CIC	λλG	TTT Lys>
101				020			1030					,			-116	LYB)
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	IGC ACC	<b>XCC</b>	CAC	XXC	AGT	CCA	CCI	TIC	CCT	GCC	000	ATC	GAG	λλλ	ACC	λTC
i	Сув	YIA	Val	λen	Ser	Ala	Ala	Pha	Pro	Ala	Pro	TAG	CTC	TIT	TGG	TAG Ile>
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•	TCC AGG			λλλ ΤΤΤ	GGC		CCG CCC	AAG		CCX	CAG	GIG		ACC	λtt	
•	TCC AGG			λλλ ΤΤΤ	GGC		CCG CCC	AAG		CCX	CAG			ACC	λtt	
•	TCC AGG Ser			AAA TIT Lys	GGC		CCG CCC	AAG	Ala	CCX	CAG GIC Gln	Val		ACC TCC Thr	ATT TAA Ile	CCA CCT Pro>
	TCC AGG Ser	Lys 10	Thr	AAA TIT Lys	GGC CCG Gly	Arg	CCG GGC Pro	AAG TTC Lys	Ala O	Siro CCY CCY	CAG GTC Gln	Val	Tyr	ACC TGG Thr	ATT TAA Ile	Pro>
3	TCC AGG Ser 11	Lys 10 ccc ccc	Thr AAG TTC	AAA TIT Lys CAG CTC	GGC CCG Gly L120 CAG	ATG	CCG GGC Pro	AAG TTC Lys 11:	Ala O GAT	CCA GCT PTO	CAG GTC Gln 11	Val	ATG Tyr	ACC TOG Thr	ATT TAA Ile L150 TGC	ATG
3	TCC AGG Ser 11	Lys 10 ccc ccc	Thr AAG TTC	AAA TIT Lys CAG CTC	GGC CCG Gly L120 CAG	ATG	CCG GGC Pro	AAG TTC Lys 11:	Ala O GAT	CCA GCT PTO	CAG GTC Gln 11	Val	ATG Tyr	ACC TOG Thr	ATT TAA Ile L150 TGC	GGT Pro>
3	TCC AGG Ser 11	Lys 10 ccc ccc	AAG TTC Lys	AAA TIT Lys CAG CTC	GGC CCG Gly L120 CAG GTC Gln	ATG	CCG GGC Pro	AAG TTC Lys 11: AAG TTC Lys	Ala O GAT	CCA GCT PTO	CAG GTC Gln 11	Val	TYT  CTG GAC Leu	ACC TOG Thr	ATT TAA Ile L150 TGC ACG Cys	ATG
	TCC AGG Ser 11 CCT CGA Pro	Lys CCC GGG Pro 116	AAG TIC Lys	AAA TIT Lys GAG CIC Glu	GGC CCG Gly L120 CAG GTC Gln	ATG TAC Met	CCC CCC CCC CCC Ala	AAG TTC Lys 111 AAG TTC Lys	GAT CTA ABP	CCA GGT PITO AAA TTT Lys	CAG GIC GID 11 GIC CAG Val	CAC Val 40 AGT TCA Ser 119	TYP CTG GAC Lett	ACC TGG Thr ACC TGG Thr	ATT TAA Ile LISO TGC ACG Cys	ATG TAC Met>
3	rec AGG Ser 11 CCT GGA Pro	Lys  CCC GGG Pro  116  ACA	AAG TTC Lys :0 • GAC	AAA TIT Lys GAG CIC Glu	GGC CCG Gly L120 CAG GTC Gln	ATG TAC Met	CCC CCC CCC CCC Ala	AAG TTC Lys 111 AAG TTC Lys	GAT CTA ABP	CCA GGT PITO AAA TTT Lys	CAG GIC GIn 11 GIC CAG Val	CAC Val 40 ACT TCA Ser 119	Tyr CTG GAC Leni	ACC TGG Thr ACC TGG Thr	ATT TAA Ile LISO TGC ACG Cys	ATG TAC Het>
3	rec AGG Ser 11 CCT GGA Pro	Lys  CCC GGG Pro  116  ACA	AAG TTC Lys :0 • GAC	AAA TIT Lys GAG CIC Glu	GGC CCG Gly L120 CAG GTC Gln	ATG TAC Met	CCC CCC CCC CCC Ala	AAG TTC Lys 111 AAG TTC Lys	GAT CTA ABP	CCA GGT PITO AAA TTT Lys	CAG GIC GIn 11 GIC CAG Val	CAC Val 40 ACT TCA Ser 119	Tyr CTG GAC Leni	ACC TGG Thr ACC TGG Thr	ATT TAA Ile LISO TGC ACG Cys	ATG TAC Met>
3	TCC AGG Ser 11 CCT GGA Pro	Lys 10 CCC GGG Pro 116 ACA TGT Thr	AAG TTC Lys 60 CAC CTG Asp	AAA TIT Lys GAG CIC Glu TIC AAG Phe	CAG GTC GIN 1120 TTC AAG Phe	ATG TAC Het 170 CCT GGA PTO	CCC GCC PFO CCC CCG Ala CTT Glu	AAG TTC Lys 11: AAG TTC Lys GAC CTG ASP	GAT CTA ABP 180 ATT TAA Ile	CCA CCT PTO AAA TTT Lys ACT TCA Thr	CAG GIC GID III GIC CAG Val	AGT TCA Ser 115 CAG CTC Glu	TYT  CTG GAC Lett  10  TGG ACC TTP	ACC TGG Thr ACC TGG Thr CAG GTC Gln	ATT TAA Ile IISO Cys IZ	ATG TAC Het>  AAT TTA ASn>
3	TCC AGG Ser 11 CCT GGA Pro ATA FAT Lle	Lys 10 CCC GGG Pro 116 ACA TGT Thr	AAG TIC Lys 60 CAC CTG Asp	CAG CTC Glu TTC AAG Phe	CAG GIV CAG GIC GID TTC AAG Phe	ATG TAC Het 170 CCT GGA PTO 122	CCC GCC Pro	AAG TTC Lys 111 AAG TTC Lys GAC CTG ASP	GAT CTA ABP LIBO ATT TAA Ile	CCA CGT PID AAA TIT LyE ACT TCA Thr	CAG GIC GID 11 GIC CAG Val	CAC Val 40 AGT TCA Ser 115 CAC CTC Glu	TYT  CTG GAC Lett  10  TGG ACC TTP	ACC TGG Thr ACC TGG Thr CAG GTC Gln	ATT TAA 11e 150 TGC ACG Cys 12 TGG ACC TTD	ATG TAC Het>
3	TCC AGG Ser 11 CCT CGA Pro ATA TAT Lle	Lys 10 CCC GGG Pro 116 ACA TGT Thr	AAG TTC Lys  GAC CTG Asp  210 CCA GGT	AAA TIT Lys GAG CTC Glu TTC AAG Phe	GGC CCG Gly L120 CAG GTC GIn TTC AAG Phe	ATG TAC Met 170 CCT GGA PTO 122	CCC GCC Pro	AAG CTG Lys 111 AAG TTG Lys GAC CTG Asp	GAT CTA ABP 180 ATT TAA Ile	CCA GGT PTO AAAA TTTT TCA TCA TAT	CAG GIC GID CIC CAG Val	LAC Val 40 AGT TCA Ser 115 GAG CTC Glu	TYT  CTG GAC Lett  10  TGG ACC TTP  240  ATC	ACC TGG Thr ACC TGG Thr CAG GTC Gln	ATT TAA IIe II50 ACC ACC Trp	ATG TAC Het> AAT TTA ABn>
	TCC AGG Ser 11 CCT GGA Pro ATA TAT LLe	Lys 10 CCC GGG Pro 116 ACA TGT Thr	AAG TTC Lys GAC CTG Asp 210 CCA GGT Pro	CAG CTC Glu TTC AAG Phe CCC CCC Ala	GGC CCC Gly L120 CAG GTC GIn TTC AAG Phe	ATG TAC Het TO CCT GGA PTO 122 AAC TTG ABD	CCC GCC Pro GCC CCG Ala GAA CTT Glu TAC ATG Tyr	AAG CTG Lys 111 AAG TTG Lys GAC CTG Asp	GAT CTA ABP 180 ATT TAA Ile	CCA GGT PTO AAAA TTTT TCA TCA TAT	CAG GIC GID CIC CAG Val	LAC Val 40 AGT TCA Ser 115 GAG CTC Glu	TYT  CTG GAC Lett  10  TGG ACC TTP  240  ATC	ACC TGG Thr ACC TGG Thr CAG GTC Gln	ATT TAA IIe II50 ACC ACC Trp	ATG TAC Het>
1250	TCC AGG Ser  11 CCT GGA Pro  ATA FAT CIT CCT GGA Pro  ATA CAT CIT CCT CCT GGA Pro  ATA CAT CIT CCT CCT CCT CCT CCT CCT CCT CCT CC	Lys 110 CCC GGG Pro 116 ACA TGT Thr CAG GTC Gln	AAG TTC Lys 60 CTG Asp 210 CCA GGT Pro	AAA TITT Lys GAG CTC Glu TTC AAG Phe GCG CCC AAla	GGC CCG Gly L120 CAG GTC GIn TTC AAG Phe CAG CTC GIu	ATG TAC Met 170 CCT GGA PTO 122 AAC TTG ABD	CCC CCC CCC Ala CTT TAC ATG Tyr	AAG TTC Lys 111 AAG TTC Lys GAC CTG AAG TTC Lys	GAT CTA AMP LIBO ATT TAA TIG AAC TTG AMB	CCA CGT PTO AAA TTT Lys ACT TCA TTCA TTCA TTCA TTCA	CAG GIC GIR GIC CAG Val CAG GIC GIR CAG GIC GIR	CAC Val  AGT TCA Ser 115 CAC CTC Glu CCC GGG PTO	ATC TYF  CTC GAC Leni 10  TCG ACC TTP  240  ATC TAG TAG TAG TAG TAG TILE	ACC TGG Thr ACC TGG Thr GTG GTC GIn ATG TAC Met	ATT TAA IIe II50 ACG Cys TCG ACC TTCD GAC CTG ASP	ATG TAC Het> ATT TAT AAT TTA ABN ACA TGT Thr>
1250	TCC AGG Ser  11 CCT IGA Pro  ATA PATA PATA PLIE CCC CCC CCC CCC CCC CCC CCC CCC CCC C	Lys 110 CCC GGG Pro 116 ACA TGT Thr CAG GTC GIn	AAG TTC Lys GAC CTG Asp CCA GGT Pro 12	GAG CTC Glu  TTC AAAG Phe  GCG CCC Ala	GGC CCG Gly L120 CAG GTC Gln TTC GAAG CTC Glu	ATG TAC Met 170 CCT GGA PIO 122 AAC TTG ABD	CCC CCC ALA CTT GIU	AAG TTC Lys 111 AAG TTC Lys GAC CTG AASp AAG TTC Lys	GAT CTA AMP 1180 ATT TAA 111e 12 AAC TTG AMB	CCA CGT Pro AAA TTT Lys ACT TGA Thr TGA Thr	CAG GIC GIR CAG Val	CAC Val  AGT TCA Ser 115 CAC CTC Glu CCC GGG PTO	ATC TYT CTC GAC Leni 10 . TCG ACC TTP 240 . ATC TAG Ile	ACC TGG Thr ACC TGG Thr CAG GTC Gln ATG TAC Met	ATT TAA IIe LISO ACG Cys TCG ACC TTp GAC CTG ASp	ATG TAC Het> OO AAT TTA AGN> ACA TCT TTIT>
1250	TCC AGG Ser  11 CCT CGA Pro  ATA TAT CIT CCG CCC CCC CCC CCC CCC CCC CCC CCC CC	Lys 110 CCC GGG Pro 116 ACA TGT Thr CAG GTC GIn GGC CCG	AAG TTC Lys GAC GCT GCT Pro	GAG CTC Glu  TTC AAG Phe  GCC CCC Ala  260	GGC CCC Gly L120 CAG GTC GIn TTC AAG Phe CAG CTC Glu	ATG TAC Met TO CCT GGA PTO 122 AAC TTG ABB	CCC GGC Pro GCC CGG Ala GAA CTT Glu TAC ATG Tyr	AAG TTC Lys 111 AAG TTC Lys GAC CTG ASp AAG TTC Lys	GAT CTA ASP 180 ATT TAA IIe 12 AAC TTG ASSS	CCA CGT PTO  AAA TTT Lys  ACT TCA Thr TCA Thr TCA Thr TCA Thr TCA Thr TCA Thr TCA	CAG GIC GIR  11 GIC CAG Val  CAG GIC GIR  CAG GIC AAT	CAC Val  AGT TCA Ser 115 CAC CTC Glu CCC GCG PTO	ATC TYT CTC GAC Len TCC ACC TTP ATC TAG Ile	ACC TGG Thr ACC TGG Thr CAG GTC GIn ATG TAC Met AAG	ATT TAA IIe II50 ACG Cys III GAC ACC TTP	ATG TAC Het> OO AAT TTA AGN> ACA TCT TTIT>

## FIG. 1 D

1300	0 1310			1	320			1330			13	40			
• .			◆,			•			•				•		
TCC CAC	CCY	GGA	AAT	λCT	TTC	ACC	TGC	TCT	CTC	TΤλ	CAT	GAG	GGC	CTG	
YCC CIC	$\alpha$	CCI	TTA	TCA	λλG	TCG	λŒ	λGλ	CAC	λλΤ	GTA	CTC	CCC	GAC	
Trp Glu	λla	Gly	λæn	Thr	Phe	Thr	Сув	Ser	Val	Lou	His	Glu	Gly	Leu)	,
1350			360			131	70		4	380					
•		_	•				•		-	300			1390		
CAC AAC	CAC (	יית גיי	) CT	CIC	110	100	~	-					•		
CAC AAC		~₽3 ~V7	WC7		~~	700	CIC	100	CAC	TCT	CCI	CCT	λλλ	TG A	rc
GTG TTG	24-	31 - 31V	160	23	TIC	100	CEAL	ACC	GIG	YCY	CCA	CCY	TIT	AC T	NG
His Asn	HIR I	17.8	THE	GIU	гЛя	ser	Leu	Ser	His	Ser	Pro	Gly	Lys:	•	
1400		1	410			142	10		14	130		1	440		
•			•				•			•					
CCA GTG	TCC 1	LIC.	GλG	CCC	TCT	CCT	CCI	λCλ	GGA	CTC	TGA	CXC	CTA	نيب	
GGT CAC	AGG 7	NAC .	CIC	CCC	λGλ	CCY	CCY	TGT	CCT	CYC	ACT	CIC	CAT	CGA	
145	o o		14	60		1	470			148	10				
	•			•			•				•				
CCY CCC	CIC.	CI.	GTA	Τλλ	λτλ	λλG	CXC	CCX	CCA	CIC	CCT	TGG	ACC	C	
CCT CCC	CAG (	CX (	CAT	λTT	TAT	TTC	CTC	CCT	CCT	GAC	GGA	) CC	TCC	Č	
														•	

5/41

**Nucleotides** 

Sequence of the murine TF8-5G9 light chain cDNA with protein translation. The essential regions of the cDNA are as follows:

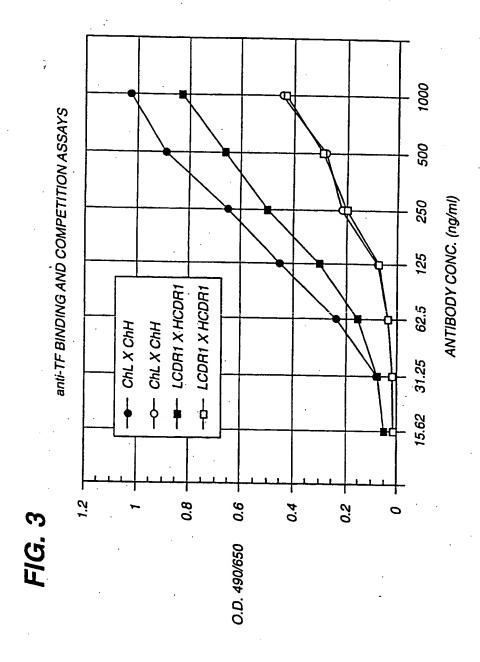
Region

			•			, COL	<u> </u>				91011						
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r I	G. 2	: A	·		5-64	4				Sta	art co	don	and	leade	er se	quenc	ce.
					65-3	385			•	Vai	riable	reg	ion.				
					386	-706		•		Mu	rine	kapp	a co	nstar	nt red	gion.	
	:				707	-917							ed re			<b>J</b>	
						-937					ly A t			9.0	• .		
	•				0.0	00.					., , , , ,	<b>W</b> 11.					
Sec	quence	Rar	ige:	1 to	937												
			1	LO			20			. 30	)		4	10			
	661		~	•	~ ~	- ~	*			- C	, 	·	~	•			
	CCT	G T	AC GC	$c \propto$	C GC	$\alpha$	ia G	rc a	ע ע	LA CC	C T	ע א	CA	C CA	G AC	C AAA	, ·
	50			60			70			ε	30			90			
	CCY	CCT	ATC	λGλ	TCT	CXC	λTC	λλG	ATG	ACC	CAG	TCT	CCA	TCC	TCC	λTG	
	CCT	CCY	TAG	TCT	<b>ACA</b>	CIG	TAG	TTC	TAC	TCC	CIC	<b>AGA</b>	CCT	ACC	λGG	TAC	
	Pro	Gly	Ile	yığ	CAB	увр	Ile	Lys	Mot	Thr	Gln	Ser	Pro	Ser	Ser	Met>	
	100			13	LO •		:	120			130			14	0		
	TAT	CCY	TCC	CIC	CCX	CYC	YCY	CTC	ACT	ATC	ACT	TCT	λλG	ccc	ACT	CAG	
•	ATA	CCI	AGC	CAC	CCI	CIC	TCT	CAG	TCA	TAG	TCA	YCY	TTC	ccc	TCA	GIC Gln>	
	131	~14	SEI	rea	CIY	GIU.	YEA	VAL	THE	TTE	THE	Сув	LYB	VIE	Ser	GID	
	:	150			160	•		1	70		1	180			190		
	CAC	ATT	λGλ	λλC	TAT	TTA	λλC	TCC	TAC	CAG	CAG	λλλ	CCA	TCC	222	طحك	
Ϊ,	cic	TAA	ici	TTC	ATA	AAT	TIG	ACC	ATG	CIC	CIC	TIT	CCT	ACC	TIT	λGλ	
	Asp	Ile	γīΔ	Lys	Tyr	Leu	λsn	TIP	Tyr	Cln	Gln	Lye	Pro	TIP	Lys	Ser>	
		2	00		:	210			220			2	30			240	-
	CCI	AAG	ACC	CIG	ATC	TAT	TAT	CCA	λCλ	AGC	TTG	CCA	GAT	CCC	CTC	CCA	
	CCY	TTC	TCC	CYC	TAG	λTλ	λTλ	CCT	TCT	TCC	YYC	CCI	CIA	`ccc	CXC	CCT	
	Pro	Lys	Thr	Leu	Ile	Tyr	Tyr	Ala	Thr	Ser	Leu	λla	YBD	Cly	Val	Pro>	
			250		,	2	60		:	270		-	280				-
	TCA	λGλ	TTC	AGT	GGC	AGT	CGA	TCT	CCG	CAA	GAT	TAT	TCT	CTA	ACC	ATC	•
	AGT	TCT	λAG	TCA	CCC	TCA	CCI	λGλ	CCC	GII	CTA	λTλ	<b>AGA</b>	CAT	TCC	TAG	٠.
	Ser	yrā	Phe	Ser	Gly	Ser	Cly	Ser	Gly	Gln	Yab	Tyr	Ser	Leu	Thr	Ile>	
	290		:	300			310			` 3:	20		:	330			
	AGC	AGC	CIG	CAG	TCT	GAC	GAT	<b>XCX</b>	GCA	ACT	TAT	TAC	TGT	CTA	CXX	CAT	
	TCG	TCC	CYC	CIC	ycy	CIC	CTA	TCT	CCI	TCX	λTλ	λTG	λCλ	CAT	CTT	GEA	
	Ser	Ser	Leu	Glu	Ser	увр	увъ	Thr	Yla	Thr	Tyr	Tyr	CAR	Lou	Gln	His>	
						-											

## FIG. 2B

	340			3	50		;	360			370			31	80	
	CCT	GAĠ	) CC	·	ma		mm-0	*	~~~		•				_	
	~~		AGC TCG		A'IT :	447.	AAG	[4.44	7	7		- TOTAL	~~~		_	
	Gly	Glu	Ser	Pro	Tyr	Thr	Phe	Gly	Gly	Gly	Thr	Lvs	Len	CIT	TAT	TTG Aun>
					-			_				-,-	~~~	GIU	116	AND
		390			400			4:	LO		4	120			430	
	)CC	CCAL	Cam	CCT	cm	003	.~	~~~	*			•			•	
	TCC	CCA	GAT CTA	CGA	CCT	CCA	TCA	CAT	ACC	TAC	TTC	CCX	ωx	TCC	ACT	CYC
	λrg	λla	λвр	λla	λla	Pro	Thr	Val	Ser	Tle	Phe	GGT.	GGT	AGG	TCA	CIC Glu>
												110	110	SEI	Ser	GIA>
		44	10		4	150			460			47	0		4	80
	CAG	Tella.	ACA.	متحكت	CCA			man.	~~~				•			•
	GTC	λλΤ	ACA TGT	AGA	CCT	CCX	CCC	AGT	CAG	CAC	TGC	TTC	TIC	χχC	YYC	TTC
	Gln	Leu	Thr	Ser	Gly	Gly	Ala	Ser	Val	Val	Cvs	Phe	Ten.	lon	TIC	AAG Phe>
	•		490				00			510				74011	VRIT	PDE)
			•				•	٠		•		•	520			
	TAC	CCC	$\overline{\lambda}\lambda\lambda$	CYC	ATC	AAT	GTC	λλG	TCC	λλG	ATT	GAT	GGC	AGT	Gλλ	CCA
	ATG	CCC	TIT	CIG	TAG	TIX	CAG	TTC	XCC	TTC	ጥአአ	(1)	~	TV-3	~	
	TYE	PTO	LYB	Авр	He	Asn	Val	Lys	TTP	Lys	Ile	yab	Gly	Ser	Clu	Arg>
53	0		:	540			550			56	50			570		
	•			•			•				•		_	•		•
	CYY	AAT	ccc	CIC	CIG	YYC	AGT	TCC	ACT	GAT	CXC	CYC	AGC	λλλ	GXC	λGC
	Gln	Agn	CCC	Val	Tan	TIG	TCA	ACC	TCA	CIA	GTC	CIC	TCC	TIT	CIC	TCG Ser>
			~~,	****	Dea	VETT	3CL	עניי	IIII	VRD	CID	ABD	ser	Lys	YBD	Ser>
	280			59	90	٠	(	500			610			62	20	
	300	TAC	100	377	*			*			•				•	
	TCG	λTG	AGC TCG	TAC	TOG	100	TGG	GAG.	TYPE	ANC	TGG	AAG	GAC	CXC	TAT	CYY
	Thr	Tyr	Ser	Met	Ser	Ser	The	Lou	Thr	Lou	Thr	Lys	VED.	Glu	TVT	Glu>
		30											-		-,-	
	•	•			640			65	•		6	60			670	
	Œλ	CAT	λλC	AGC	TAT	ACC	TCT	CAG	GCC	ACT	cxc	λλG	ACA	TCA	عرت	TCA
	CCT	GIX	TIG	TO	λTλ	TCC	<b>XCX</b>	CTC	$\infty$	TGA	GTG	Jalk	77.7	ACT	TCA	3 (***)
	VLA	His	Yed	Ser	TYT	Thr	CAR	Glu	Ala	Thr	Hie	Lys	The	Ser	Thr	Ser>
		68	10			590			700			71	0		-	20
			•			•			•				•			'20 *
	222	ATT	CIC	AAG	YCC	TIC	YYC	YCC	λλΤ	CYC	<b>TCT</b>	TA C	IAC I	CX A	VAG C	סדם סדם
	ماماما	TAA	CAC	TIC	TCC	XXC	TIC	TCC	TŢÀ	CIC	y <sub>C</sub> y	AT (	י סד	CI 1	TC C	AG GAC
	110	110	Val	_y=	Sel	PDO	ABD	VIÀ	APD	GIU	Cya	•				
		73	0	•	•	740			750			76	0		7	70
			•		<u>.</u> .	•			•				•			•
	TCT	CCC	CXC	CYC	CAG	CIC	CCC	AGC	TCC	ATC	CIX	TCT	TCC	CII	CTA	AGG
			CIC	410	410	نالما	فاجات	فللد	فاضم	فللانا	CAT	Mix	AGG	CYY	GAT	TCC
			780			75	90		1	300			810			
		<b>~~</b>	•				•			•	٠					
	AGA	YCC	<b>AGG</b>	CIT	CCC	CVC	λλG	CCY	CCI	YCC	<b>ACT</b>	CIT	CCC	CLC	CTC	CXX
		-	TCC	www	غاماما	GTG.	TIC	CT	بالقاقا	166	JCY.	CYY	œc	CYC	CYC	CIT

## FIG. 2 C



RECTIFIED SHEET (RULE 91)
ISA/EP

### FIG. 4 A

The pEe6TF8HCDR20 expression vector DNA sequence. The coding regions of the TF8-5G9 CDR-grafted HC gene, TF8HCDR20, are translated.

Sequence Range: 1 to 7073

		•	10			20			30				40		
Gλλ	TTC	GCC	GCC	ACC	ATG	GAA	TGG	AGC	TGG	CTC	Jalah	CTC	مكيف _	JAN-	
CTT	λλG	CCC	CCG	TGG	TAC	CIT.	YCC	TCG	YCC	CAG	λλλ	GAG	AAG	110	220
					Met	Glu	Trp	Ser	TIP	Val	Phe	Leu	Phe	Phe	Leu)
50 •			60			•	70 *			80			90		
TCA	GTA	ACT	ACA	CCT	GTA	CAC	TCA	CNA	GII	CAG	CTG	GTG	GAG	طحكك	CCN
AGT	CAT	TGA	TGT	CCA	CAT	GTG	AGT	GTT	CAA	37	CAC	CAC	~~~		
Ser	Val	Thr	Thr	Gly	Val	His	Ser	Gln	Val	Cln	Lau	Val	Glu	Ser	Gly>
•	00			110			120				30			L40	
GGA	CCA	GTA	GTA	CAA	CCT	GGA	λGG	TCA	CIG	λGλ	CTG	TCT	TGT	λλG	GCT
CCI	CCT	CXT	CAT	CIT	CCA	CCI	TCC	λCT	CYC	TCT	GAC	AGA	ACA	<b>Jake</b>	CCA
Gly	Gly	Val	Val	Gln	Pro	Gly	yra	Ser	Lou	yrg	Leu	Ser	Сув	Lys	Ala>
	150			10	•			L70			180				90
λGT	CCY	TTC	λλτ	ATC	AAG	GAC	TAT	TAT	ATG	CAC	TGG	GTC	λGλ	CAA	CCT
TCA	CCI	YYC	TIX	TAG	ĬΤ	CIG	λTλ	ATA	TAC	GTG	ACC	CAG	طمخك	2777	CC2
ser	CIA	Phe	Asn	Ile	Lys	YBD	Tyr	Tyr	Met	His	Trp	Val	yra	Gln	Ala>
	:	200			210			22	20 -		2	230			240
~~		•			•				•			•			•
GGA	CCA	7777	CGA	CIC	GAG	TCC	ATA	CCT	TTA	ATT	CAT	CCI	CYC	<b>AAT</b>	CCT
- Court	CCI	***			CIC	ALL	TAT	CUA	AAT	TAA	CTA	GGA	crc	TIA	CCA
Pro	Glv	I.ve	Clv	Leu	Glu	The second	TIA		T	T1 -	<b>)</b>	~	~		
Pro	Gly	Lys	Cly	Leu	Glu	TIP	Ile	CIA	Leu	Ile	ysb	Pro	Glu	λen	Cly>
PTO	GIĀ	Lys 25	20 CIÀ	Leu	Glu	TEP			270			28	30		-
AAC PTO	YCC	Lye 25 ATA	Cly TAT	CAT	CCC	TIP 260 AAG	TTC	CAA	270 GGA	λGλ	·TTC	28	30 *	المحالة	CCA
AAC	ACG	Lys 2: ATA TAT	Gly  TAT  ATA	GAT CTA	CCC	TIP 260 AAG TIC	TTC AAG	CAA GTT	270 GGA CCT	AGA TCT	TTC	ACA TOT	ATT	TCT	GCA
AAC	ACG	Lys 2: ATA TAT	Gly  TAT  ATA	GAT CTA	CCC	TIP 260 AAG TIC	TTC AAG	CAA GTT	270 GGA CCT	AGA TCT	TTC	ACA TOT	ATT	TCT	CCA
AAC TTG ABD	ACC TGC Thr	25 ATA TAT Ile	TAT ATA Tyr 300	GAT CTA Asp	CCC GGG Pro	AAG TTC Lys	TTC AAG Phe	CAA GIT Gln	270 GGA CCT Gly	AGA TCT ATU	TTC AAG Phe	ACA TGT Thr	ATT TAA Ile	TCT AGA Ser	GCA CGT Ala>
AAC TTG Asn 290	ACG TGC Thr	Lys 2: ATA TAT Ile	GIY  TAT  ATA  TYE  300	GAT CTA Asp	CCC GGG PTO	AAG TTC Lys	TTC AAG Phe	CAA GIT Gln	270 GGA CCT Gly	AGA TCT ATU	TTC AAG Phe	ACA TGT Thr	ATT TAA Ile	TCT AGA Ser	GCA CGT Ala>
AAC TIG ABN	ACG TGC Thr	Z: ATA TAT Ile TCT AGA	TAT ATA TYF 300 AAG	GAT CTA Asp	CCC GGG PTO ACA TGT	AAG TTC Lys	TTC AAG	CAA GTT Gln CTG	270 GGA CCT Gly CAG	AGA TCT ATU 320 ATG	TTC AAG Phe GAC	ACA TGT Thr TCA	ATT TAA Ile 330 CTC GMG	TCT AGA Ser	GCA CGT Ala>
AAC TIG ABN	ACG TGC Thr	Z: ATA TAT Ile TCT AGA	TAT ATA TYF 300 AAG	GAT CTA Asp	CCC GGG PTO ACA TGT	AAG TTC Lys	TTC AAG	CAA GTT Gln CTG	270 GGA CCT Gly CAG	AGA TCT ATU 320 ATG	TTC AAG Phe GAC	ACA TGT Thr TCA	ATT TAA Ile 330 CTC GMG	TCT AGA Ser	GCA CGT Ala>
AAC TIG ABN	ACG TGC Thr AAC TTG ABD	Z: ATA TAT Ile TCT AGA	TAT ATA Tyr 300 AAG TTC Lys	GAT CTA Asp	CCC GGG PTO ACA TGT	AAG TTC Lys	TTC AAG	CAA GTT Gln CTG	270 GGA CCT Gly CAG	AGA TCT ATU 320 ATG	TTC AAG Phe GAC CTG Asp	ACA TGT Thr TCA	ATT TAA Ile 330 CTC GAG Leu	TCT AGA Ser	GCA CGT Ala>
AAC TTG ABD GAC CTG ABD GAG	ACG TGC Thr AAC TTG Asn	Lys 2: ATA TAT Ile TCT AGA Ser	TAT ATA TYR 300 AAG TTC Lys	GAT CTA ABD AAT TTA ABD	CCC GGG Pro ACA TGT Thr	AAG TTC Lys 31 CTG GAC Leu	TTC AAG Phe AAG Phe 360	CAA GTT Gln CTG GAC Leu	270 CGA CCT Gly CAG GTC Gln	AGA TOT ATU 320 ATC TAC Met 37	TTC AAG Phe GAC CTG ABP	ACA TOT Thr TCA AGT Ser	ATT TAA Ile 330 CTC GAG Leu	TCT AGA Ser AGA TCT ATG	GCA CGT Ala> CCT GGA Pro>
AAC TTG ABD GAC CTG ABD GAG CTC	ACG TGC Thr AAC TTG ASD	Lys 2: ATA TAT Ile TCT AGA Ser ACA TGT	TAT ATA TYT 300 AAG TIC LYB	GAT CTA ASP AAT TTA ASB GTC CAG	CCC GGG PTO ACA TGT Thr	AAG TTC Lys 31 CTG GAC Leu	TTC AAG Phe  TTC AAG Phe  360  TGT ACA	CAA GIT Gln CIG GAC Leu GCT	270 GGA CCT Gly CAG GTC Gln	AGA TOT ATU 320 ATG TAC Met 37 GAT	CAC CTG AMP	ACA TOT Thr TCA AGT Ser	ATT TAA Ile 330 CTC GAG Leu TAT	TCT AGA Ser AGA TCT AIT B0 TAC	GCA CGT Ala> CCT GGA Pro>
AAC TTG ABD GAC CTG ABD GAG CTC	ACG TGC Thr AAC TTG ASD	Lys 2: ATA TAT Ile TCT AGA Ser ACA TGT	TAT ATA TYT 300 AAG TIC LYB	GAT CTA ASP AAT TTA ASB GTC CAG	CCC GGG PTO ACA TGT Thr	AAG TTC Lys 31 CTG GAC Leu	TTC AAG Phe  TTC AAG Phe  360  TGT ACA	CAA GIT Gln CIG GAC Leu GCT	270 GGA CCT Gly CAG GTC Gln	AGA TOT ATU 320 ATG TAC Met 37 GAT	CAC CTG AMP	ACA TOT Thr TCA AGT Ser	ATT TAA Ile 330 CTC GAG Leu TAT	TCT AGA Ser AGA TCT AIT B0 TAC	GCA CGT Ala> CCT GGA Pro>
AAC TIG ABD 290 GAC CIG ABD GAG CIC GIU	ACG TGC Thr AAC TTG ASn O GAT CTA Asp 390	Z: ATA TAT Ile TCT AGA Ser ACA TGT Thr	GIY  TAT  ATA  TYT  300  AAG  TTC  Lys  GCA  CAT  Alla	GAT CTA ABP AAT TTA ABB GTC CAG Val	CCC GGG Pro  ACA TGT Thr  TAC ATG Tyr	TIP 260 AAG TYC Lys 31 CTG GAC Leu TAT TYT	TTC AAG Phe 360 . TGT ACA Cys	CAA GIT GIN CIG GAC Leu GCT CGA Ala	270 GGA CCT Gly CAG GTC Gln AGA TCT ATY	AGA TCT ATG 320 ATG TAC Met 37 GAT CTA ABP	TTC AAG Phe GAC CTG ASP AAC TTG ASR 420	ACA TGT Thr TCA AGT Ser AGT TCA Ser	ATT TAA IIe 3330 . CTC CAG Leu TAT ATA ATA TYT	TCT AGA Ser AGA TCT Ary ATG TAC ATG TYF	GCA CGT Ala> CCT GGA PTD> TTC AAG Phe>
AAC TIG ABD 290 GAC CIG ABD GAG GAG GAG GAC GAC	ACG TGC Thr AAC TTG ASn GAT CTA Asp 390 TAC	Z: ATA TAT Ile TCT AGA Ser ACA TGT Thr	GIY  TAT  ATA  TYT  300  AAG  TTC  Lys  GCA  CCT  Ala	GAT CIA ABD AAT TIA ABD GTC CAG Val	CCC GGG Pro  ACA TGT Thr  TAC ATG Tyr  GGA	TIP 260 AAG TIC Lys 31 CTG GAC Leu TAT TYT ACA	TTC AAG Phe 10 TTC AAG Phe 360 TGT ACA Cys	CAA GIT GIR CTG GAC Leu CCT CCA Ala	270 GGA CCT Gly CAG GTC Gln AGA TCT ATY	AGA TCT ATU 320 ATC TAC Met 37 GAT CTA ABP	TTC AAG Phe GAC CTG AMP  70 AAC TTG AMR 420 AGC	ACA TOT Thr TCA AGT Ser ACT TCA Ser	ATT TAA 11e 330 . CTC CAG Leu TAT ATA TYF	TCT AGA SET AGA TCT ATG ATG TTAC ATG TYT	GCA CGT Ala> CCT GGA PTO> TTC AAG Phe>
AAC TTG ABD CTG GAG CTG GAU GAC CTG	ACC Thr AAC TRO Thr AAC TRO Asn IO ATTO CTA Asp 390 TAC ATG	Z: ATA TAT Ile TCT AGA TGT TTH TGG ACC	GIY  TAT  ATA  TYT  300  AAG  TTC  Lys  GCA  CGT  Ala  GGCC  CCC  CCCC	GAT CTA ASP AAT TTA ASP GTC CAG Val	CCC GGG Pro  ACA TOT Thr  TAC ATG TYT  GGA CCT	ACA TOT	TTC AAG Phe  TTC AAG Phe  ACA Cys	CAA GIT GIR CIG GAC Leu CCT CCA Ala	270 GGA CCT Gly CAG GTC GIn AGA TCT ATY	AGA TCT ATU 320 ATC TAC Met 37 GAT CTA Asp	TTC AAG Phe GAC CTG AMP	ACA TOT Thr TCA AGT Ser TCA Ser	ATT TAA 330 . CTC CAG Leu TAT ATA TYT	TCT AGA SET AGA TCT ATG ATG TYT	GCA CGT Ala> CCT GGA PTO> TTC AAG Phe>

## FIG. 4 B

440	450 *	46	0 470 •	480
ANG GGC CCA	TCC GTC TTC	ccc cre ece	CCC TGC TCC AGG A	GC ACC TCC
TTC CCG GGT	AGG CAG AAG	GGG GXC CCC	GGG ACG AGG TCC T	CC TCC ACC
Dyn Gly Flo	ser val Phe	Pro Deu Ala	Pro Cys Ser Arg S	er Thr Ser>
490	n 50	10 5	520	
		₩.	*	
GAG AGC ACA	SCC GCC CTG C	CC TGC CTG C	TC AAG GAC TAC TO	NG GGG CTT
Clu Ser Thr	Ala Ala Leu (	ly Cys Leu V	al Lys Asp Tyr Pl	he Pro Glu>
Δ.		550	¥.	70
•	540 *	•	•	•
CCC GTG ACG	CTC TCC TCC	NAC TCA GGC	CC CTG ACC AGC G	SC GTG CAC
GGC CAC TGC	CAC AGC ACC!	MED SET CLU (	CG GAC TGG TCG C	ly Val His>
PIO VAI IM	var ser rip.	•		
580	590 *	. 600	610	620 *
ACC TTC CCG	CCT CTC CTA	CAG TOC TCA	CCA CTC TAC TCC C	TC AGC AGC
TGG AAG GGC	CGA CAG GAT	GTC AGG AGT	CCT GAG ATG AGG G Gly Lou Tyr Ser L	AG TOG TOG ou Ser Ser>
Thr Phe Pro	VIS AST DOG	om ser ser		
630 .	640	650	. 660	670
GTG GTG ACC	GTG CCC TCC	AGC AGC TTG	GGC ACG AAG ACC T	AC ACC TGC
CAC CAC TGG	CAC GGG AGG	TCG TCG AAC	CCC TCC TTC TCC A	ITG TGG ACG
Val Val Thr	Val Pro Ser	Ser Ser Leu	Gly Thr Lys Thr T	yr ine cyso
680	690	70	0 710	720 • •
AAC GTA GAT	CAC AAG CCC	AGC AAC ACC	ANG GTG GAC ANG A	CA GIT GGT
TITG CAT CITA	CTC TTC GGG	TCG TTG TGG	TTC CAC CTG TTC T Lys Val Asp Lys J	CL CYY CCY
Asn Val Asp	HIS LAS PLO	Ser Am Tar	THE VAL ABO DIE A	TY VALV
73	30 7	740	750 760	0 · ◆
GAG AGG CCA	GCX CXC GGC	AGG GAG GGT	GTC TGC TGG AAG	CCA GGC TCA
CTC TCC CCT	CCT CTC CCC	ACC CAC CCY	CAG ACG ACC TTC	GGT CCG AGT
770	780	790	800	810
פטר נער נער •	CCT GGA CGC	ACC CCG GCT	CTC CAG CCC CAG	CCC AGG GCA
SEC CYC CYC	CCA CCT CCC	TCG CCC CCA	CAC GTC GGG GTC	GGG TCC CGT
820	830	840	850	860
•	•	•	•	**************************************
CCA AGG CAT CGT TCC GTA	CCC CCX TCT	CAG AGG AGT	CCC CCY CCC CLC	ACT GGT GGG
870	880	890	900	910
•	•	•	•	•
CAC TCA TGC	TCA GGG AGA	GGG TCT TCT	CCT ANA ANG GTG	CAG GCT CCG

## FIG. 4 C

	920		930			94	10			950			960
CCC ACC	CAC ACC	CTG CAC	GAT CTA	CCC	CCT GGA	ACC TCC	CCY	ccc	CCT CCA	000 000	CAT GTA	ACA TGT	CCC
970		980			990			1000					
GCA GGT CGT CCA	CCY CCC	CTC	AGA TCT	CCT GGA	CCC	AAG TTC	agc TCG	C'AT GTA	atc Tag	CCC	CIC	CTG	CCT GGA
1010 1020			1030			1040				1050			
CCC CCT	CIC CAN	AGC	CCA CCT	CCC	CAA GTT	AGG TCC	CCA	AAC	TCT AGA	CCA GCT	CIC	CCI	CXC
1060 10		.070	1		F080		1090		11		.00		
CTC AGA GAG TCT	CAC CT	CIC	TCC	TCC AGG	CAG	<b>λ</b> ΥΥ Τ <b>λλ</b>	CCA GCT	CTA CAT	ACT TGA	CCC	AAT TTA	CII	CTC GAG
1110	10 11		20 1			130			1140			1150	
TCT GCA AGA CGT	CTC ACC	TİT	λTλ	CCA	CCC	CCT	ACC	CCT	<b>AGT</b>	ACG	CCT	CCA	AAG TTC
1	160	1	170			11	₿0		1:	190		:	1200
CCA ACC	CAC CC	TCG	CCC GGG	TCC	AGC TCG	TCA AGT	AGG TCC	CCC	GAC	AGG	TGC	CCT	λGλ
	1210		1220				1230			1240			
CAT CCC	TGC AT	CAG	CCT CCT	CAG	CCC	CCA CCT	CCC	CCC	TGC ACG	TGA ACT	000 000	ATC TAG	CAC GTG
1250	126	)		127	70		:	1280			129	0	
CTC CAT	CTC TT	CTC	AGC TCG	TC	CA C	IC Y	AG G	AC C	<b>20 01</b>	er c	GT A	er c	TC TTC AG AAG al Phe
1300	1	310		1	320			1330			13	40	
CTG TTC GAC AAG Lou Phe	CCC CC	TIT T	CCC	TTC	CIG	TGX	CAG	TAC	TAG	ACC	GCC	TCG	GGA
1350		1360	•		13	70		1	380			1390	٠,
כדכ כאנ	ACC TC TCC AC	CYC S	CYC	CXC	CIG	CAC	TCG	CTC	CIT	CIG	CCC	CTC	CAG
14	100	1	410			1420	)		14	30		1	440
GTC AAC	AAC TG TTG AC AAD Tr	C ATG	CAC	CTA	. CCC	CAC	CTC	CAC	GTA	TTA	CCG	TTC	TGT

### FIG. 4 D

1450 1460 1470 1480 ANG CCG CCC GAG GAG CAG TTC AND AGO ACG TAC CCT CTG CTC AGO CTC THE COE COE CHE CHE GHE ANG THE TOE TOE ATC COA CAE CAG TOE CAG Lys Pro Arg Glu Glu Gln Phe Asn Ser Thr Tyr Arg Val Val Ser Val> 1490 1500 1510 1520 CTC ACC GTC CTG CAC CAG GAC TGG CTG AAC GGC AAG GAG TAC AAG TGC GAG TGG CAG GAC GTG GTC CTG ACC GAC TTG CCG TTC CTC ATG TTC ACG Leu Thr Val Leu His Gln Asp Trp Leu Asn Gly Lys Glu Tyr Lys Cys> 1540 1550 1560 AMG GTC TCC AMC AMA GGC CTC CCG TCC TCC ATC GAG AMA ACC ATC TCC TTC CAG AGG TTG TIT CCG GAG GGC AGG AGG TAG CTC TIT TGG TAG AGG Lys Val Ser Asn Lys Gly Leu Pro Ser Ser Ile Glu Lys Thr Ile Ser> 1600 1610 1620 AAA GCC AAA GG TGG GAC CCA CGG GGT GCG AGG GCC ACA TGG ACA GAG GTC TIT CGG TIT CC ACC CTG GGT GCC CCA CGC TCC CGG TGT ACC TGT CTC CAG Lys Ala Lys> 1640 1650 1660 1670 1680 AGO TOG GOO CAO COT CTG COO TGG GAG TGA COG CTG TGC CAA COT CTG TOG AGO COG GTG GGA GAO GGG ACO CTC ACT GGC GAO ACG GTT GGA GAO 1690 1700 1710 1720 1730 TCC CTA CA GGG CAG CCC CGA GAG CCA CAG GTG TAC ACC CTG CCC CCA TCC AGG CAT GT CCC GTC GGG GCT CTC GGT GTC CAC ATG TGG GAC GGG GGT AGG Gly Gln Pro Arg Glu Pro Gln Val Tyr Thr Leu Pro Pro Ser> 1740 1750 1760 1770 1780 CAG GAG GAG ATG ACC AAG AAC CAG GTC AGC CTG ACC TGC CTG GTC AAA GTC CTC CTC TAC TGG TTC TTG GTC CAG TGG CAC TGG ACG CAC CAG TTT Gln Glu Glu Met Thr Lys Asn Gln Val Ser Leu Thr Cys Leu Val Lys> 1800 1810 GGC TTC TAC CCC AGC GAC ATC GCC GTG GAG TGG GAG AGC AAT GGG CAG CCC AAG ATG GGG TCG CTG TAG CGG CAC CTC ACC CTC TCG TTA CCC GTC Gly Phe Tyr Pro Ser Asp Ile Ala Val Glu Trp Glu Ser Asn Gly Gln) 1830 1840 1850 1860 CCC GAG AAC AAC TAC AAG ACC ACG CCT CCC GTG CTG GAC TCC GAC GGC GGC CTC TTG TTG ATG TTC TGG TGC GGA GGG CAC GAC CTG AGG CTG CCG Pro Glu Asn Asn Tyr Lys Thr Thr Pro Pro Val Leu Asp Ser Asp Gly> 1880 1890 1900 1910 1920 TCC TTC TTC CTC TAC AGC AGG CTA ACC GTG GAC AAG AGC AGG TGG CAG AGG AAG AAG GAG ATC TCC TCC GAT TGG CAC CTG TTC TCG TCC ACC GTC Ser Phe Phe Leu Tyr Ser Arg Leu Thr Val Asp Lys Ser Arg Trp Gln>

## FIG. 4 E

1	930		;	1940			19	50		1	960			1970	
CIC	CCC	TTA	CAG	λλG	AGT	λCG	λGG	CAC	TAC	CIX	CTC	CCA	GAC	GTG	AAC TTG AED>
	198	30		19	990			2000			201	LO		2	2020
CIG	ATG	TGT	GTC	TTC	AGC TCG Ser	GAG	λCC	GAC	YCY	GAC	CCX	TIT	A CT	C AC	E CA
	20	30		2	2040			205	50 •		20	60		2	1070
CCC	CCC	CAA GTT	CCC	CCC	CCT CCA	CCC	000 000	GCT CGA	CTC GAG	CCC	CTC CAG	œc	CCY CCY	GGA CCT	TGC ACG
		201	30		20	90		2	2100			21.1	10		
TTG AAC	GCA CCT	CCT CCA	ACC TGG	CCC	TCT AGA	ACA TGT	TAC ATG	TTC AAG	CCA GGT	CCC	ACC TGG	CAG GTC	CAT GTA	CCI	AAT TTA
2120	•		2130			214	•			L50 +			2160		•
AAA TTT	CCI	CCC	YCC TCC	ACT TGA	CCC	CTG GAC	CCC	CCC	TGT ACA	CIC	XCT TGX	CAC	ATG TAC	CTT CLA	CIT CAA
217	70 +		23	180		2	2190			220	00		22	210	
TCC AGG	XCG TGC	CCX	CAG	CCC	CIC	TCT AGA	GAG CTC	CCC CCC	TGA ACT	CXC	aca Tgt	TGA ACT	CCC	AGG TCC	CYC
2	2220			22:	30		22	40		2	2250	٠		226	0
AGC TCG	000 000	TCC AGG	CAC GTG	TGT ACA	CCC	CAC	XCT TGX	ccc	cci ccy	CCC	TCT ACA	CCX CCT	CCY	CTC CAC	CCT GGA
	22	70		2	2280			229	0		23	00		2	310
CCC CCC	CCY	CCY	AGG TCC	CXC	CCC	CYC	AGC TCG	CIC	CCC	CIC CIC	CCC	TCG AGC	CCA CCT	CCC	TGG ACC
		232	20		23	330		2	2340		·	235	50		
CCC	ATT TAA	TGC ACG	CXC	CCI	ccc	CCI	CCC	TCC AGG	AGC TCG	AGC TCG	AGG TCC	XCT TGX	CTA GAT	CIC	GAT CTA
2360	·	2	2370			238	30		2.	90		:	2400		
CXT	AAT TTA	CIC	CCX	TAC ATG	CAC	λΤΤ Τλλ	TCT ACA	YCY YCY	GCT CCA	TTT AAA	ACT TGA	TGC ACG	TTT AAA	AAA TTT	AAA TTT
241	LO .		24	20		2	2430			244	0		24	150	
CCI	CCC	ACA TCT	CCI	CCC	CCY CCI	GAA CTT	CCI	GAA CTT	ACA TGT	TAA ATT	AAT TTA	CAA CTT	TGC ACG	AAT TTA	TCT

## FIG. 4 F

2460	,		24	•			480			2490				00
ACA ACI	KKŤ T	CIT	CXX CXX	TAT <b>AT</b> A	TGC	AGC TCG	ΤΤ <b>λ</b>	TAA ATT	TGG	TTA AAT	CYY	<b>ATA</b>	AAG	CAA
2	510		:	2520			25	30	•	2	540			2550
TAG CAT ATC GT	CYC	AAA TTT	TTT	CAC	AAA TTT	TAA ATT	AGC TCG	ATT	TTT	TTC	ACT	CCY	TTC	TAG
	256				70			2580			259		~~	, i
		•			•			•				•		
ANC NCC	TTT	CAG	CAA GTT	ACT TGA	CAT GTA	CAA GTT	TCT ACA	ATC TAG	TTA AAT	TCA AGT	TCT ACA	CIC CIC	CIA	CCI
2600	2	2610			262	20		26	30		2	2640		
CTA CGC	CCC	XCG TGC	CAT GTA	CCT CCA	CCC	CCC	CAT GTA	CAC GTG	CCC	သာ သာ	CAC GTG	AGG TCC	TGC	CCA
2650			60			670			268			. 26		
	~~~			~~~	-	~~	ma	~~~	m^^	-	101		•	
TGC TGG ACG ACC														
2700			271	.0		27	20		2	730			274	•
CCA CTT	CCC	CCT	CAT	GAG	CCC	TTG	TIT	œc	CCT	CCC	TAT	CCT	CCC	λGG
GGT GAA														
2	750 •	٠.	2	760			277	70 +		27	780		:	2790
CCC CTC	CCC	CCC	CCI	CIG	TTG AAC	ccc	CCC	ATC TAG	TCC AGG	TTG AAC	CXT GTX	CCA CCT	CCX	TTC AAG
•	280	00		28	110		:	2820			283	30		
CTT CCC	CCC	CCC	GTG	CTC	AAC	CCC	CTC	AAC	CTA	CTA	CTC	CCC	ACT.	مخلعة
CAN CCC														
2940	:	2850			286	50 •		21	370 •		:	2880		
CTA ATO														
2890		2	900		:	2910			29:	20		2	930	
													•	
CYC CCC														
2940			29	50		2	960		:	2970 •			29	80
CCI CCI														

## FIG. 4 G

	2	990			3000			30	10		3	020			3030
CJ/C	600 000	TTT	ccc	CCT	CCI	AGC TCG	TCC	CTC	GIG	CCC	TCT	CCI	CIT	. ccc	ACC
		30				050			3060			30			166
CTG	ccc	CIT	ACC	GGA	TAC	CTG	TCC	GCC	Jelel.	بالت	ىلىك	T***	•	100	~~~
CAC	GGC	GAA	TCC	CCT	ATG	GAC	AGG	CCC	λλλ	CXC	CCX	AGC	CCI	TOS	CAC
3080			3090			31	00		3	110			3120		
CCC CCC	CII	TCT AGA	CAA	TGC	TCA AGT	CCC	TGT ACA	AGG TCC	TAT ATA	CIC	AGT TCA	TCG	CYC	TAG ATC	CIC
	30 .			140			3150			31				170	
CTT CAA	CCC	TCC	AAG TTC	CIG	CCC	TGT ACA	GTG CAC	CAC	GAA CTT	ccc	CCC	CAA	CAG	CCC	GAC
	3180			31	90		3:	200		:	3210			32	20
323 323	TGC	CCC	TTA AAT	TCC	CCA	AAC TTG	Τ <b>λ</b> Τ <b>λ</b> Τλ	CCI	CIT	CAC CTC	TCC	AAC	CCC	GTA CAT	AGA TCT
		230			3240			32				260			3270
CAC	GAC	TTA	TCG	CCA	CIG	GCA	GCA	CCC	ACT	CCT	λλC	AGG	ידידג		•
GIG	CTG	AAT	AGC	GGT	GAC	CCT	CIT	œc	TCA	CCY	TIC	TCC	Tλλ	TOC	TCT
		328	0		32	90		3	300			331	.0		
323 323	AGG TCC	TAT ATA	CAT	CCC	CCX	GCT.	ACA TGT	CIC	TTC AAG	TTG AAC	AAG TTC	TGG ACC	TGG ACC	CCI	AAC TTG
3320		3	330			334	, o		33	50		3	360		
TAC ATG	CCC	TAC ATG	XCT TGA	AGA TCT	AGG TCC	ACA TGT	GTA CAT	TTT AAA	CCA	ATC TAG	TGC	GCT CGA	CIC	CTG GAC	AAG TTC
337				80			390			340				10	
CCX	CII	<b>XCC</b>	TIC	CCX	λλλ	λGλ	CIT	GGT	AGC	TCT	TGA	TCC	GGC	<b>XXX</b>	CAA .
GGT	CAA	TCG	λλG	CCI	TTT	TCT	CNA	CCX	TCC	YĊY	λCT	AGG	CCG	TIT	CIT
3	420			343	•		34	40		3	450			346	0
ACC TGG	ACC TGG	CCY	CCY	AGC TCG	CCY	CCX	TTT AAA	TTT AAA	CYY	YCC TCC	XXG TTC	CXG GTC	CXG CXG	ÄTT TAA	ACC TGC
	34	70		. 3	480			349	0		35	00		. 3	510
သေ	AGA TCT	XXX TTT	XXX TTT	GGA CCT	TCT	CAA	CII	GAT CTA	cci.	TTG AAC	ATC TAG	TTT	TCT AGA	ACC TGC	ece cec

## FIG. 4 H

		35	20		3	1530			3540	)		35	550		
74.77			•			. •			•	•			•		
YCY	כבע	CCA	GIC	ACC	TTC	CIT	TTC	YC	, cc.	יגד ז גאז	, cc.	S AT	TTC	CAC	ATG TAC
3560			3570				80			590			3600		
AGA TCI	TTA	TCA	AAA TTT	AGG	ATC	TIC	ACC	TAG	TAG	CIT	TT	A A A T	TA	, XX	TGA
_	10			620			•						. VI.	111	ACT
	•			•			3630				40			650	
act TCA	XXX XXX	TIT	TCA AGT	TAG	TAA ATT	) TCX	`	TAT ATA	CIC	TAX ATI	ACT	TGC	TCT	CTC	) TCA
	3660	,		36	•			680				_			00
TAC ATG	CAN	YCC YCC	TTA	ATC TAG	AGT TCA	CIC	CCA	CCI	ATC TAG	TCA	. ccc	ATC TAG	TGT	CTA	TTT
		710			3720			37				740			3750
CCT CCA	TCX AGT	TCC AGG	<b>λΤλ</b> ΤλΤ	CAA CAA	ccc	TGA ACT	CTC GAG	CCC	~	CYC	TAG	ATA TAT	ACT TGA	ACC TCC	λΤλ ΤΑΤ
		37				770			3780			37		-	
CCC	CAG	GGC	TTA	CCA	TCT	CCC	ccc	λGΤ	CCT	GCA	λTG	λΤλ	œ	CCA	GAC
	CIC	فكك	AAT	GGT	λGλ	CCC	GGG	TCA	CCA	CCT	TAC	TAT	CCC	CCI	CIG
3800			3810			38:	20		3	830			3840		
CCA	CCC	TCA	CCC	CCT	CCX	CAT	TTA	TCA	GCA	λτλ	AAC	CAG	ccy	GCC	GGA
385				CCA	GGI			Mer	CGT	TAT	TIC	CIC	CCT	ccc	CCT
	•			•.			3870			381	•			B90	
AGG TCC	CCC	CIC	<b>ccc</b> ccc	AGA TCT	AGT TCA	CCY CCY	CCI	CCI.	ACT TGA	TTX <b>λλ</b> Τ		CCC	TCC AGG	ATC TAG	CAG
	900			391				20			930			394	
TCT	ATT	AAT	TGT	TGC	* CCG	Gλλ	CCT	AGA	GTA	ACT	•	<b>~~~</b>	~~		•
AGA	TAA	TTA	ХСХ	ACG	CCC	CTT	CGA	TCT	CAT	TCA	TCA	AGC	CCL	CAA	XXT TTX
	39	50		. 3	960			397	0		39	80		. з	990
AGT	TTC	œc	AAC	CII	CII.	CCC	λΤΤ	CCT	γςγ	GCC	እጥሮ	•	CIV		•
TCA	λλC	ထေ	TIC	CAA	CYY	œc	TAA	CCY	TCT	0	TAG	CXC	CXC	AGT	ccc
		400	0		40	10		4	020			403	0		
TCC	TCC	TTT	CCT	λTG	CCT	TCA	TTC	) AGC	ىلىك +	مضی	<b>~~~</b>	<b>.</b>	•		
AGC	λGC	λλλ	CCA	TAC	œλ	AGT	λλG	TO	λGG	CCY.	YCC	CIT	CCI	TCA AGT	agg TCC

## FIG. 4 I

4040		4	050			406	50		40	70	•	4	1080		
CCT	CAA CAA	ACA TGT	TGA ACT	TCC AGG	CCC	ATG TAC	TTG AAC	TGC ACG	AAA TTT	AAA TTT	000 000	CTT CAA	AGC TCG	TCC AGG	TTC AAG
409	0		43	00		•	1110			413	20		41	130	
CCY	CCT	CCC	ATC TAG	CAA	CXC	AGA TCT	XCT TCX	XXG TTC	TTG AAC	CCC CCC	CCX	CYC	TTA AAT	TCA AGT	CIC CIC
•	140			415	50		4:	160		4	170			418	30
ATG TAC	CXX	ATG TAC	CCA CCT	CCX CCT	CTG	CXT CTX	AAT TTA	TCT AGA	CIT	ACT TGA	CXG	ATG TAC	CCA CCT	TCC AGG	CTA CAT
	4:	190		•	<b>£</b> 200			42:	10		4:	220		•	1230
λGλ TCT	TGC	TTT	TCT	ĠTG CAC	ACT	GGT	CIC	TAC	TCA	ACC	λλG	TCA	TTC	TGA	CYY
		424				250			4260			42		<i></i>	
TAG	TCT	ATC	• ccc	CCA	ccc	Agt	TCC	TCT	TGC	œ	ccc	TCA	· ACA	œ	CAT
ATC	λCλ	TAC	ccc	CCI	CCC			YCY			œc	AGT	TCT	ccc	CTA
4280		4	1290			430	90		43	10		4	1320		
_										_					
λλΤ	ACC TGG	CCC	CCA GGT	CAT GTA	AGC TCG	aga TCT	act Tga	TTA AAT	AAA TTT	CIC	CTC CXG	ATC TAG	λΤΤ Τλλ	CCT	AAA TTT
λλΤ	TGG	CCC	CCT	CAT GTA	AGC	TCT	ACT TGA 1350	TTA AAT	AAA TŢT	GTG CAC	CYC	ATC TAG	TAA	GGA CCT	AAA TTT
AAT TTA 43:	TGG	TCG	GGT 43 GGG	GTA 340 CGA	TCG	TCT	TCA 1350 TCA	AAT AGG	TŢT ATC	436 TTA	CAG	TAG	TAA 43	CCT 70	TCC
AAT TTA 43: CCT GCA	TGG 30 TCT AGA	CGC	GGT 43 GGG	GTA 340 CGA GCT	AAA TTT	TCT	TCA TCA AGT	AGG TCC	TŢT ATC	436 TTA AAT	CCC CCC CCC	TAG	TAA 43	CCT	TCC
AAT TTA 43: CGT GCA	TGG 30 TCT AGA 1380	TCG AGC	GGT 43 GGG CCC	GTA  340  CGA  GCT  439	AAA TTT	CTC	TGA 1350 TCA AGT	AGG TCC	ATC TAG	TTA AAT	CAG CCG CGC	TAG CTG GAC	TAA 43 TTG AAC	CCT 70 AGA TCT 442	TTT TCC AGG
AAT TTA 433 CGT GCA AGT	TGG TCT AGA 1380 TCG	TCG AGC	GGT 43 GGG CCC	GTA  440  CGA  GCT  439	AAA TTT	CTC	TCA 1350 TCA AGT 44	AGG TCC	ATC TAG	TTA AAT	CAG CCG CGC 1410	TAG CTC GAC	TAA 43 TTG AAC	CCT 70 AGA TCT 442	TCC AGG
AAT TTA 433 CGT GCA AGT	TGG 10 TCT AGA 1380 TCG AGC	TCG AGC	GGT 43 GGG CCC	GTA  GGA  GCT  439  CCCC  GCC	AAA TTT	CTC	TCA 1350 TCA AGT 44	AGG TCC	ATC TAG AAC TTG	TTA AAT	CAG 60 CCC GGC 1410 TCT AGA	TAG CTC GAC	TAA 43 TTG AAC	AGA TCT 442 TCT AGA	TCC AGG
AAT TTA 43: CGT GCA AGT TCA	TGG TCT AGA 1380 TCG AGC	TCG AGC ATG TAC	GGT 43 GGG CCC TAA ATT	GTA  GGA  GCT  439  CCC  GGG	AAA TTT 90 ACT TGA	CTC GAG CGT GCA	TCA 1350 TCA AGT 44 GCA CCT	AGG TCC 100 CCC GGG	ATC TAG	TTA AAT TGA ACT	CAG CCG GGC 1410 TCT AGA	CTG GAC TCA AGT	TAA  TTG AAC  GCA CCT	AGA TCT 442 TCT AGA	TTT TCC AGG
AAT TTA 43: COT GCA ACT TCA	TGG TGT AGA 1380 TGG AGC	TCG AGC	GGT 43 GGG CCC TAA ATT	GTA GGA GCT 435 CCC GGG	AAA TTT 90 ACT TGA	CTC GAG	TCA 1350 TCA AGT 44 GCA CGT	AAT AGG TCC 100 CCC GGG 44:	ATC TAG	TTA AAT TGA ACT	CAG CCC CCC CCC CCC A110 TCT ACA CCA	TAG CTG GAC TCA AGT	TAA  43  TTC AAC  GCA CCT	AGA TCT 442 TCT AGA	TTT TCC AGG
AAT TTA 43: COT GCA ACT TCA	TGG TGT AGA 1380 TGG AGC	TCG AGC ATG TAC 130 ACC TCG	GGT 43 GGG CCC TAA ATT	GTA GGA GCT 435 CCC GGG	AAA TTT 90 ACT TGA 4440 TCT AGA	CTC GAG	TCA 1350 TCA AGT 44 GCA CGT	AAT AGG TCC 100 CCC GGG 441 GCA CGT	ATC TAG	TTA AAT TGA ACT	CAG CCC CCC CCC CCC A110 TCT ACA CCA	TAG CTG GAC TCA AGT	TAA 43 TTG AAC GCA GCT CMA GTT	AGA TCT 442 TCT AGA	TTT TCC AGG
AAT TTA 433 CGT GCA AGT TCA ACT TCA	TGG TGT AGA 1380 TCG AGC TTC AGC	TCG AGC TAC TGG AGC TGG	GGT 43 GGG CCC TAA ATT AGC TCG	GTA  GGA  GCT  CCC  GGG  GTT  CAA	AAA TTT 90 ACT TGA 4440 TCT AGA	CTC GAG CGCA CGCA CGCA CGCA CGCA CGCA CGCA	TCA AGT  TCA AGT  TCA AGT  AGA  AGA  TCA ACA	AAT AGG TCC 400 CCC GGG 441 GCA CGT	ATC TAG  AAC TTG  AAA  TTT  4500	TTA AAT TGA ACT ACT	CAG CCG CGC L410 TCT AGA CCT TGA	TAG CTG GAC TCA AGT AGG TCC 45:	TAA 43 TTG AAC GCA CCT CAA CCTT	CCT 70 AGA TCT 442 TCT AGA AAT TTA	TTT TCC AGG TTTT AAA 4470 GCC CGG
AAT TTA 433 CGT GCA AGT TCA ACT TCA	TGG TGT AGA 1380 TCG AGC TTC AGC	ATG TAC TGG AAG TTC	GGT 43 GGG CCC TAA ATT AGC TCG	GTA  GGA  GCT  CCC  GGG  GTT  CAA	AAA TTT 90 ACT TGA 4440 TCT AGA	CTC GAG  CGT GCA  GGG GCC  GGG GCC  GGG GCG GCG	TCA AGT  TCA AGT  TCA AGT  AGA  AGA  TCA ACA	AAT AGG TCC LOO CCC GGG 441 GCA CGT	ATC TAG  AAC TTG  AAA TTT  AAA TTT	TTA AAT TGA ACT ACT	CAG CCG CGC L410 TCT AGA CCT TGA	TAG CTG GAC TCA AGT AGG TCC 453	TAA 43 TTG AAC GCA CCT CAA CCT CTC	CCT 70 AGA TCT 442 TCT AGA AAT TTA	TTT TCC AGG 30 TTTT AAA 470 GCC CGG
AAT TTA 43: CGT GCA AGT TCA ACT TEA 4520 TTC	TGG TGT AGA 1380 TCG AGC AGC AGC ATT AAA TTT CTT	ATG TAC TGG AAG TTC	GGG CCC  TAA ATT  AGC TCG  GGA CCT  4530	GTA  GGA  GGCT  439  CCCC  GGG  GTT  CAA  ATA  TAT	AAA TIT  O  ACT TGA  4440 TCT AGA AGG TCC	CTC CAG CCCA CCCCA CCCCA CCCCA CCCCCA CCCCCA CCCCCA CCCCCA CCCCCC	TCA ACT TCA ACT TCA ACT TCA ACT ACA ACT ACA ACA	AAT AGG TCC 100 CCCC GCG 441 GCA CGT CGG GCC	ATC TAG  AAC TTG  SO  AAA TTT  4:  TAT	TTA AAT TGA ACA TGT ACA TGT CAC	GAG  CCG GGC  1410 TCT AGA  4  GGA CCT  TGA ACT	TAG CTC GAC TCA AGT 450 AGG TCC 451 ATA TAT	TAA 43 TTG AAC GCA GCT CAA GTT CTC GAG 4560	TCT A42 TCT AGA AAT TTA ATA TAT	TTT TCC AGG TTT AAA 470 GCC CGG

## FIG. 4 J

	45	70		4:	580		•	4590			46	00		4	510	
	agc TCG	CCT	TAC ATG	<b>XTX TXT</b>	TTT	CTT	TCT	ATT TAA	TAG ATC	XXX TTT	AAT TTA	AAA TTT	CYY	λΤ <b>λ</b> ΤλΤ	CCC	CAA
-	,	4620			46	30		4	640			4650			466	50
	000 000	000 000	ACA TCT	TTT	ccc	CCA	AAA TTT	GTG CAC	CCY	CCT	CTG	CAG	TAA ATT	CAA	ACC TCG	ATT
			670			4680			46				700			710
	ATT	λTY	ATC	)C)	747.2	•	Th in			•	~~				ccc	•
	TAA	TAG	TAC	TGT	AAT	TCC	ATA	TTT	TTA	TCC	CCA	TAG	TGC	TCC	ccc	TGA ACT
			473	•			730			4740			47	•		
	TGG	CTC	TTT	ccc	CCA	ccc	ATC	CIT	CCI	AAT	GIT	000	TCC	CXC	CCA CCT	GGA
						-			•••	***	حمم		ACC	GIG	GCT	CCT
47	60		. •	1770			47	B0		4	790	•		4800		
	CYY	ccc	TCA	AGA	CYY	AAT	CTA	ATC	λCλ	CTG	CCT	CAC	CIT	œ	CTG	CCC
	CTT	GGG	AGT	ıcı	CII	TTA	CAT	TAG	TGT	GAC	CCA	CIC	CAA	CCC	CYC	CCC
	48:	•			B20 •			4830			48	•			850	·
	CIT	TCT	CCC	TIT	λτλ	AGG	YCY	CXC	TIT	ATG	TTT	λλG	λλG	CIT	CCT	λλλ
	w.x	المغالم	age	***	TAT	TCC	TCT	GIG	λλλ	TAC	λλλ	TTC	TTC	CYY	CCY	TTT
		4860			48	70		41	980			4890			490	10
	TTC	CIT	GCG	GCT	TTG	GCA	GCC	λλG	(ALA)	GAG	3700	*			GTG	•
,	λλG	GAA	CGC	CGA	AAC	CCT	CCC	TTC	GAT	CIC	TAG	AGA	TOG	λλG	CAC	TCA AGT
		4	910	•	4	1920			493	30		49	940		4	950
	λCC	ACG	GTG	ACT	GCA	GTG	λλΤ	λλΤ	λλλ	ATG	TGT	ململت	ىلتىل. ب	~~	λλλ	*
	TCC	TGC	CAC	TGA	CCT	CAC	TTA	TTA	TIT	TAC	λCλ	CAA	λCλ	GGC	TIT	ATG
			496	0		45	70		- 4	980			499	0	•	
	ccc	TTT	TGA	GAT	TTC	TGT	CCC.	CCA	CTA	AAT	TCA	TYPE	ccc	ecc.	λτλ	~~~
(	CCC	λλλ	λCT	CTA	λλG	УСУ	GCG	CCT	CAT	TTA	AGT	λCλ	ထေ	œc	XTX TXT	CAC
50	00		5	010			502	20		50	30		5	040		
	TG	Jalab	ATC	GCC +	CATT	101	C3 M	•	~~ ·		•					
(	CAC	λλλ	TAG	CCC	CTA	TCT	CTA	000	CTA	TAA	CCI	XXX TTT	XTC TAG	CIA CIA	ATT TAA	TGA ACT
	505	50		50	060		:	5070			50	80		5	090	
	λλλ	TAT	GGC	λΤλ	TTG	λλλ	λTG	TCG	ccc	λTG	TGA	GIT	TCT	GTG	TAA	Carc
	TTT	ATA	ccc	TAT	λλC	TTT	TAC	AGC	GGC	TAC	λCT	CYY	λCλ	CYC	ATT	CYC

## FIG. 4 K

5100		5110	51	120	5130	5140
ATA TOG	CCA TIT	TTC CAA	TTC ACT	TIT TIG	GGC ATA CC	C GAT ATC TGG G CTA TAG ACC
51	150	5160		5170	5180	5190
CGA TAG	CGC TTA	TAT CCT	TTA CCG	GGG ATG	GCC ATA CA	C GAC TIT GGT
GCT ATC				•		CTG AAA CCA
	5200	Э.	210	5220	<b>&gt;</b>	230 •
CLC YYC	CCC CTA	TCT GTG	YCY CCC	$\lambda\lambda\lambda$ $T\lambda T$ $TTT$ $\lambda T\lambda$	CCC ACT TT	C CAT ATA GGT G CTA TAT CCA
5240	5250		5260	5:	270	5280
GAC AGA CTG TCT	CGA TAT	CTC CCA	ATA TOG TAT AGO	CCG ATA	CAG CCC AC	A TCA AGC TGG T AGT TCG ACC
5290	· 5	300.	5310		5320	5330
CAC ATG	GCC AAT	CCT ATA	CCA TCT	ATA CAT	TGA ATC AA	T ATT CGC CAT
5340		- 5350	5	360	5370	5380
TAG CCA	TAT TAT	TCA TTG	GTT ATA	TAG CAT	AAA TCA AT	A TTG GCT ATT
ATC GGT	XTX XTX	ACT AAC	CAA TAT	ATC GTA	TTT AGT TA	T AAC CGA TAA
5:	390	5400		5410	5420	5430
GGC CAT	TGC ATA	CCT TCT	ATC CAT	ATC ATA	ATA TOT AC	TANA TAT AAC
						I AAA TAT AAC
	5440		•	. 5460		470
GCT CAT CGA GTA	CAG GTT	CAT TAC	CCC CAT	CAY CLC	ATT GAT TA TAA CTA AT	T TGA CTA GTT A ACT GAT CAA
5480	5490		5500	5	510	5520
λττ λλτ	AGT AAT	CAA TTA	CCG GGT	CAT TAG	TTC ATA GC	C CAT ATA TGG
TAA TTA	TCA TTA	GTT AAT	ecc ccy	GIA ATC	ANG TAT CO	G GTA TAT ACC
5530	_	540	5550 •		5560 *	5570
AGT TCC TCA AGG	GCG TTA	CAT AAC GTA TTG	TTA CCC AAT GCC	TAA ATG ATT TAC	CCC CCC CI	C CCY CLC CCC
5580		•	56	•	5610	5620
CCA ACG	ACC CCC TGG GGG	GCC CAT	TCA CCT ACT GCA	CAA TAA GTT ATT	TGA CCT AT	G TTC CCA TAG C AAG GGT ATC

## FIG. 4 L

		56	530		•	640			565	50		56	60		5	670
						CTT										
			568	30		56	90		5	700		,	571	0		
						TGG ACC										
5720	)		:	5730			574	0		57	750		5	760		
						TCX AGT										
5	77	0		57	780		:	5790			580	00		58	110	
						<b>T</b> እፐ እፐእ										
	5	820			583	30		56	340		!	5850			586	50
						TTA AAT										
		5	870		:	5880			589	90		5	900		:	5910
						CCA										
			59	20		5	930			5940		•	59	50		
																ACT TGA
5960	0			5970 •			59	80	•	5	990			6000	٠	
																CTA CAT
6	501	.0		6	020		(	6030			604	.0	•	60	50	
						AGG TCC										
`	6	060			60	70 •	-	6	080		4	090			610	00
						GAC CTG										
*		6	110			6120			61:	30	•	6:	L40 •		(	5150
<b>8</b>	NC TG	ACC TGG	CCC	ACC TGG	CIX	CCA	CCC	TCC	CCC	CCC	CCC	AAC TTG	CCA	CCA CCT	TTG AAC	CII

### FIG. 4 M

6160 6170 6180 6190 CCC GGA TTC CCC GTG CCA AGA GTG ACG TAA GTA CCC CCT ATA GAG TCT GCG CCT AMG GGG CAC GGT TCT CAC TGC ATT CAT GGC GGA TAT CTC AGA 6210 6220 6230 ATA GGC CCA CCC CCT TGG CTT CTT ATG CAT GCT ATA CTG TTT TTG GCT THE CCC GGT GGG GGA ACC GAA GAA TAC GTA CGA THE GAC ANA AAC CGA 6270 6250 6260 TGG GGT CTA TAC ACC CCC GCT TCC TCA TGT TAT AGG TGA TGG TAT AGC ACC CCA GAT ATG TGG GGG CGA AGG AGT ACA ATA TCC ACT ACC ATA TCG 6300 6320 6340 6310 TTA GCC TAT AGG TGT GGG TTA TTG ACC ATT ATT GAC CAC TCC CCT ATT ANT CCG ATA TCC ACA CCC ANT ANC TCG TAN TAN CTG GTG AGG GGA TAN 6370 GGT CAC GAT ACT TTC CAT TAC TAA TCC ATA ACA TGG CTC TIT GCC ACA CCA CTG CTA TGA AAG GTA ATG ATT AGG TAT TGT ACC GAG AAA CGG TGT 6420 6430 6410 ACT CTC TIT ATT GGC TAT ATG CCA ATA CAC TGT CCT TCA GAG ACT GAC TGA GAG AAA TAA CCC ATA TAC GGT TAT GTG ACA GGA AGT CTC TGA CTG 6440 6450 ACC GAC TOT GTA TIT TTA CAG GAT GGG GTC TCA TIT ATT ATT TAC AAA TGC CTG AGA CAT AAA AAT GTC CTA CCC CAG AGT AAA TAA TAA ATG TTT 6500 6510 6490 TTC ACA TAT ACA ACA CCA CCG TCC CCA GTG CCC GCA GTT TTT ATT AAA AMG TOT ATA TOT TOT GOT GGC AGG GGT CAC GGG CCT CAA AMA TAA TTT 6550 6560 6570 6580 6540 CAT AAC GTG GGA TCT CCA CGC GAA TCT CGG GTA CGT GTT CCG GAC ATG GTA TIG CAC CCT AGA GGT GGG CIT AGA GCC CAT GCA CAA GGC CIG TAC 6600 GGC TOT TOT COG GTA GOG GGG GAG CTT CTA CAT COG AGC COT GCT CCC CCG AGA AGA GGC CAT CGC CGC CTC GAA GAT GTA GGC TCG GGA CGA GGG ATG CCT CCA GCG ACT CAT GGT CGC TCG GCA GCT CCT TGC TCC TAA CAG TAC GGA GGT CGC TGA GTA CCA GGG AGC CGT CGA GGA ACG AGG ATT GTC

#### **RECTIFIED SHEET (RULE 91)**

## FIG. 4 N

6680		6	690			670	0		67	710		6	720		
466	ACG	CCA	GAC	TTA	CCC	λĊλ	GCX	CCA	TGC	$\alpha$	$\infty$	CCA	CCX	GTG	TGC
ACC	TCC	GCT	CIG	AAT	ccc	TCT	CCT	CCT	ACG	CCT	CCT	CCI	CCT	CYC	ACG
673	30		67	40	,	6	750			676	0		67	70	
CGC	λCλ	AGG	222	TGG	œc	TAG ATC	GCT	ATG	TGT	CTC	λλλ	ATG	AGC	TCG	GGG
ထော	TGT	TCC	CCC	ACC	GCC	ATC	منم	TAC	<b>NUN</b>		111	TAC	1146	MIC.	a.
•	5780			675	0		68	300		(	5810			682	20
100	-	Calab	CCA	ccc	<u></u>	λCG	CAT	TTG	GAA	GAC	TTA	λCG	CAG	CCG	CAG
TCG	CCC	CYY	CT	CCC	CYC	TGC	GIA	AAC	CII	CIG	AAT	TCC	CIC	GCC	CTC
Ť	6	830		(	5840		*	68	50		6	860		. (	6870
NG	116	ATC	CAG	GC)	CCL	GAG	Jale:	<b>1777</b>	TCT	TCT	GAT	AAG	ACT	CAG	3.00
						CTC									
		68	B0		6	B90 •		,	6900	٠.		69	10		
TAA	CTC	ccc	TTG	œ	TCC	TCT	TAA	CCG	TGG	λGG	GCX	CTC	TAG	TCT	GAG
λTT	CXC	GGC	λλC	CCC	λŒ	YCY	ATT	CCC	ACC	TCC	CCI	CYC	ATC	ycy	CIC
6920			6930			69	40	•	6	950			6960		-
.020			-		~~~	~~	•	~~1	~~~	~			-		
															YCI TCI
69	70		6	980			6990			70	00		7	010	
(T)	ACA	GAC	ىلتىك	4CC	Jefet		TCC	ريت	لملمك	. 44-ai	CCN	CTC	300	بالملتا	CTT
															GAA
	7020			70	30		7	040			7050	) ,		70	60
															ccc
CIG	TGC	TIC	CAA	ccc	GAC	CTC	CVC	CIN	CCI	. CYC	ATC	TCC	TAG	CIN	CCC
	7	070			+			٠							• .
ccc	GCC	AGC	TC												
ĠΦ	CCC	TCG	AG.												

### FIG. 5 A

The pEe12TF8LCDR3 expression vector DNA sequence. The coding regions of the TF8-5G9 CDR-grafted LC gene, TF8LCDR3, are translated.

Sequence Range: 1 to 7864 50 AAT TOA OC ATG GGT GTG COA ACT CAG GTA TTA GGA TTA CTG CTG CTG TGG TTA AGT GG TAC CCA CAC GGT TGA GTC CAT AAT CCT AAT GAC GAC GAC ACC Met Gly Val Pro Thr Gln Val Leu Gly Leu Leu Leu Trp> 60 70 CTT ACA GAT GCA AGA TGT GAT ATC CAA ATG ACA CAA TCT CCT TCT TCT GAA TOT CTA COT TCT ACA CTA TAG GTT TAC TGT GTT AGA GGA AGA AGA Leu Thr Asp Ala Ary Cys Asp Ile Gln Met Thr Gln Ser Pro Ser Ser> 100 110 120 CTA AGT GCT TCT GTC GGA GAT AGA GTA ACA ATT ACA TGT AAG GCG AGT CAT TOA COLA AGA CAG COT CTA TOT CAT TOT TAA TOT ACA TTO COO TOA Leu Ser Ala Ser Val Gly Asp Arg Val Thr Ile Thr Cys Lys Ala Ser> 150 160 170 CAG GAC ATT AGA AAG TAT TTA AAC TGG TAT CAG CAA AAA CCT GGG AAG GTC CTG TAA TCT TTC ATA AAT TTG ACC ATA GTC GTT TTT GGA CCC TTC Gin Asp Ile Arg Lys Tyr Leu Asn Trp Tyr Gin Gin Lys Pro Gly Lys> 200 210 220 230 GCT CCT AMG CTA CTG ATT TAT TAT GCA ACA AGT TTG GCA GAT GGA GTA CCA GGA TTC CAT GAC TAA ATA ATA CCT TCT TCA AAC CCT CTA CCT CAT Ala Pro Lys Leu Leu Ile Tyr Tyr Ala Thr Ser Leu Ala Asp Gly Val> 260 270 290 CCT TCT AGA TIT TCT GGT TCT GGC TCT GGA ACA GAC TAC ACA TTC ACA GGA AGA TOT AAA AGA COA AGA COG AGA COT TOT CTG ATG TOT AAG TOT Pro Ser Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Tyr Thr Phe Thr> 300 310 320 ATT TOT TOT CTC CAA COT GAG GAC ATT GOT ACA TAC TAC TGC CTA CAA TAA AGA AGA GAG GIT GGA CTC CTG TAA CGA TGT ATG ATG ACG GAT GIT Ile Ser Ser Leu Gln Pro Glu Asp Ile Ala Thr Tyr Tyr Cys Leu Gln> 340 350 360 380 CAT GGT GAG AGT CCC TAT ACA, TTT GGA CAA GGA ACA AAA CTA GAG ATC GTA CCA CTC TCA GGC ATA TGT AAA CCT GTT CCT TGT TTT GAT CTC TAG His Gly Glu Ser Pro Tyr Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile> 390 400 ACA AGA ACT GIT GOG GOG COG TOT GIC TIC ATC TIC COG COA TOT GAT TGT TGT TGA CAA CGC CGC GGC AGA CAG AAG TAG AAG GGC GGT AGA CTA

## RECTIFIED SHEET (RULE 91) ISA/EP

Thr Ary Thr Val Ala Ala Pro Ser Val Phe Ile Phe Pro Pro Ser Asp>

## FIG. 5 B

	440			45	0		4	60			470			48	10
GAG	CAG	TTG	λλλ	TCT	GGA .	ACT	GCC	TCT	GTT	CTG	TGC	CTG	CTG	AAT	* A A C
CIC	GTC	AAC	TTT	λGλ	CCI '	TGA	CCC	λGλ	CAA	CAC	λCG	GAC	CAC	Colots	-
Glu	Gln	Leu	Lys	Ser	Gly	Thr	λla	Ser	Val	Val	Сув	Leu	Leu	λsn	Yes:>
	•	90			500			51	0		5	520			530
TTC	TAT	ccc	λGλ	CAG	GCC	λλλ	GTA	CXG	TGG	λλG	CIC	CAT	λλC	CCC	CTC
λAG	λTλ	CCC	TCT	CIC	CCC	TTT	CAT	GTC	YCC	TTC	CXC	CIX	TTG	ccc	GAG
Phe	Tyr	Pro	yrg	Glu	Ala	Lys	vai	GIH	TIP	гав	٧٨١	ивр	VRII	VTS	Leu>
			40		_	50			560				70		
CAA	TĆG	GCT	λλC	TCC	CAG	GAG	AGT	GTC	YCY	CAG	CAG	GYC	YCC	AAG	GAC
GIT	AGC	CCY	TTG	ACC	GTC	CTC	TCA	CAG	TGT	CTC	CIC	CIG	TCG	TTC	yab>
Gln	Ser	GTĀ	VBII	Ser	GIN	GIU	SEL	VAL	1111	GIU	GIII	למע	SEI	ryn	VRD
580 +			590 •			, -	•			610			620		
AGC	ACC	TAC	AGC	CIC	AGC	YCC	YCC.	CTG	λCG	CIG	AGC	λλλ	€CX	CYC	TAC
TCG	TCC	ATG	TCG	CAC	TCC	TCG	TCC	GAC	TGC	GAC	TOS	TAT	Ala	CIG	ATG Tyr>
Ser	Thr	TYT	SEI	Leu	367	SET	2111	nea				27.0	~~~	nap	IYL
	30			640			650 •	•			60 •	٠		670	
GAG	λλλ	CYC	λλλ	CIC	TAC	GCC	TCC	CYY	CTC	ACC	CAT	CAG	CCC	CIG	AGC
CTC	TTT	GIG	TIT	CAG Val	ATG	112	VCC	Glu	Val	TOC	Hig	GID	Glv	LAC	Ser>
GIL	Ly =	***	wy-	***	-3-	,	c, s						,		5527
	680			6	90		,	700		,	710			7	20
TCG	ccc	CTC	λCλ	λλG	AGC	TTC	AAC	λCC	CCA	GAG	TCT	TA	CY C	CC Y	ga agt
													CI C	CC T	CI TCA
Ser	Pro	Val	Thr	Lys	Ser	Phe	Yen	YIA	GIY	Glu	СУВ	,	•		•
		30			740			750 •			-	60			770
GCC	CCC	, YCC	TGC	TCC	TCA	CTI	CCY	GCC	TCC	CCY	TC	TAA	TCA	GCC	λΤλ
ccc	CCC	TCC	: ACG	AGG	AGT	CXX	GGT	CCC	ACC	CCI	Mai	. Ald	, Me.	ناخِکا :	TAT
		780	• .			90			800			810	•		
CCA	CAT	TIC	TAG	, AGG	TTT	TAC	TIC	CTT	יאד :	* ***	, ACC	TC	CX	. YCC	TCC
CCI	CI	<b>, yy</b> (	) ATC	: TCC	: λλλ	ATC	XXC	: ເນ	\ XII	TT	TCC	S ACC	GR	TCC	λCC
820			830			840	)			350			960		
•			•			•	•			•			•		
ccc	TC	/ YC	TC	AAC	XTX	XX	( TC)	XX	CÝ.	TIC	TR	TI	TT	A AC	r TGT
CCC	: AC	r TC	; AC	TIC	TAI	TT	r AC	C TAC	: GI	r yy(	: AN	: AX	∴ <b>XX</b> ′	r TC	A ACA
870	)		1	380			890			900	)			910	
	• •	,		•			•			•	•	•		•	
TT	A II	C CX	cr	T AT	A ATC	GT	r AC	<b>A A</b> A'	T AA	A GC	A AT	X CC	A TC	y Cy	A ATT
AX:	L YY	CCI	C CY	A TA	r TAC	CY	A TC	r TT	A TT	r cg	r TX	TC	I YC	r cr	T TAA

### RECTIFIED SHEET (RULE 91)

## FIG. 5 C

	9	20			930			9	40		9	50			960	
	TCX	CXX	ÄTA	AAG	CAT	TTT	TTT	CAC	TGC	ATT	CTA	GTT	CTG	GIT	TGT	CCA
	λGΤ	CIT	TAT	TTC	CTA	λλλ	λλλ	CIC	YCC	TÄÄ	CAT	CYY	CYC	CYY	YCY	CCT
		. 97	70 *		9	80			990			100	0		1	010
	λλC	TCA	TCA	ATG	TAT	CIT	ATC	ATG	TCT	GGA	TCC	TCT	λCG	CCC	GAC	GCA
	TIG	AGT	AGT	TAC	ATA	GAA	TAG	TAC	AGA	CCT	AGG	AGA	TGC	GGC	CIC	CCT
		1	020			103	0		10	340		1	.050			
	TCG	TGG	CCG	GCA	TCA	ccc	ccc	CCA	CAG	GTG	œc	TTG	CIG	CCC	CCT	λΤλ
	λGC	ACC	GGC	CGT	λGT	CCC	CCC	CCT	GIC	CAC	CCC	AAC	GAC	CCC	CCX	TAT
106	0		10	70		1	080			109	0		11	.00		7
-	*			•			•				•			•		
					CCC											
1	110			112	20		11	30		1	140			115	0	
	TCA	GCG	CIT	CTT	TCG	CCG	TGG	GTA	TGG	TGG	CAG	ccc	CCT	GGC	• ccc	ccc
					AGC											
	13	160		1	1170			111	30		, 11	.90		1	1200	
					CAT											
	TGA	CAA	ccc	CCC	GTA	CXC	GXA	CCT	λŒ	TCC	TAA	CCY	ACG	ccc	ccc	CCX
		12:	10		13	220			1230			12	10		1	250
		CAA	•		CAA	ccr		λCT	GGG			CCT	• AAT		GGA	GIC
		CAA	•			ccr		λCT	- CCC	CAC		CCT	AAT TTA		GGA	GIC
		CAA GTT	•		CAA	ccr	TCA	λCT	- CCC			CCT	• AAT		GGA	GIC
	CCA	CAA GTT	260 CCC	GGX AGX	CAA GTT	CCT GGA 12'	TGA 70 ACC	ACT TGA	GGG CCC	280 280	CIT	CCT	AAT TTA 1290 GGC	CCI	CCT	CCA
	CCA	CAA GTT	260 CCC	GGX AGX	CAA	CCT GGA 12'	TGA 70 ACC	ACT TGA	GGG CCC	280 280	CIT	CCT	AAT TTA 1290 GGC	CCI	CCT	CCA
130	CCA CCA CCT	CAA GTT TAA ATT	ccc ccc 1260 ccc	AGA TCT	CAA GIT GCG GCG	CCT GGA 12' TCG AGC	TGA  70  ACC TGG	ACT TGA TGG AGC	GGG CCC	280         	CAA CAA 30	CCT GGA GCT CGA	AAT TTA 1290 GGC CCG	CCT CAA	GGA CCT TTT AAA	CCY CCY
130	CCA CCT O	CAA GTT TAA ATT	. CCC	AGA TCT 310	CAA GTT GCG GCG	CCT GGA 12' TCG AGC	TGA  70 ACC TCG 1320 CCA	ACT TGA TGG AGC	GGG CCC 1 GGC CCC	280 280 CCC CCC 13	GAA GTT CAA	CCT GGA GCT CCA	AAT TTA 1290 GCC CCG	CCT CAA 340 CTC	GGA CCT TTT AAA	CCA
	CGA CGT 00 TAG ATC	CAA GTT TAA ATT	. CCC	AGA TCT 310 CCC	CAA GTT GCG GCG	CCT GGA 12' TCG AGC	TGA  70 ACC TGG  1320 CGA GCT	ACT TGA TGG AGC	GGG CCC 1 GGC CCC	CAC 280 CCC CCC 13 CAA CTT	GAA GTT CAA	CCT GGA GCT CGA	AAT TTA 1290 GCC CCG	CCT CAA 340 CTC GAG	GGA CCT TTT AAA	GTC CAG
	CGA CGT OO TAG ATC	CAA GTT TAA ATT GCT CGA	* CGG GGC	AGA TCT 310 CCC GGG	CAA GTT CCC CCC CCC	CCT GGA 12' TCG AGC	TGA 70 ACC TGG 1320 CGA GCT	ACT TGA TGG AGC	CCC  CCC  TCA AGT	CAA	GAA GTT CAA 30 AAA TTT	CCT GGA CCA TCG AGC	AAT TTA 1290 GGC CCG	CTT CAA 340 CTC GAG	GGA CCT TTT AAA AAG TTC	CCA CCA CCT CCA CCT
	CCA CCT TAG ATC	CAA GTT TAA ATT CCA	ccc	AGA TCT 310 CCC GGG	CAA GTT CCC CCC CCC	CCT GGA 12" TCG AGC	TGA  70  ACC TGG  1320  CGA GCT  AGG	ACT TGA TGG AGC	GGG CCCC	CAA CTT	GAA GTT CAA 30 AAA TTT 1380	CCT GGA GCT CGA TCG AGC	AAT TTA 1290 CCC CCC 1	CTT CAA	GGA CCT TTT AAA AAG TTC	GTC CAG
	CGA CGT TAG ATC	CAA GTT TAA ATT CCA	ccc	AGA TCT 310 CCC GGG	CAA GTT CCC CCC CCC	CCT GGA 12' TCG AGC TGA ACT	TGA  70  ACC TGG  1320  CGA GCT  AGG	TCG AGC GCA GCT AGT	GGG CCCC	CAA CTT	GAA GTT CAA 30 AAA TTT 1380 ATA	CCT GGA GCT CGA TCG AGC	AAT TTA 1290 CCCC CCCC 1 ACCC TCCC	CTT CAA	GGA CCT TTT AAA AAG TTC	CCA CCA CCT CCA CCT
	GCA GCA CGT TAG ATC L350 GAG CTC	CAA GTT TAA ATT CCA CAC	260 GCC 1260 GCC CCC 1. CCC GCC CCC CCC CCC CCC CCC CCC CCC CC	AGA TOT 310 CCC GGG 13 AAA TTT	CAA GIT GCC CCC CCC CCC GGG	CCT CGA ACC	TGA  70  ACC TGG  TGG  CGA  GCT  AGG  TCC	ACT TCA  TCG AGC  GCA CGT  370  ACT TCA	GGG CCC  1  GGC CCC  TCA AGT  ATA TAI  20  TGI	CAA CCC CCC CCC 13 CAA GTT	CAA CIT CAA 30 AAA TIT 1380 ATAT TAT	CCT CCA CCA AGC	AAT TTA CCCC CCC 1 ACCC CCCC CCCC CCCC C	CTT CAA 340 CTC GAG 13 CAA CTC CTC	GGA CCT TIT AAA AAG TIC	CCA CCC CCC CCC CCC CCC CCC CCC CCC CCC
	GCA GCA CGT TAG ATC L350 GAG CTC	CAA GTT TAA ATT CCA CAC	260 GCC 1260 GCC CCC 1. CCC GCC CCC CCC CCC CCC CCC CCC CCC CC	AGA TOT 310 CCC GGG 13 AAA TTT	CAA GIT GCC CCC CCC CCC GGG	CCT CGA ACC	TGA  70  ACC TGG  TGG  CGA  GCT  AGG  TCC	ACT TCA  TCG AGC  GCA CGT  370  ACT TCA	GGG CCC  1  GGC CCC  TCA AGT  ATA TAI  20  TGI	CAA CCC CCC CCC 13 CAA GTT	CAA CIT CAA 30 AAA TIT 1380 ATAT TAT	CCT CCA CCA AGC	AAT TTA CCCC CCC 1 ACCC CCCC CCCC CCCC C	CTT CAA 340 CTC GAG 13 CAA CTC CTC	GGA CCT TIT AAA AAG TIC	CCA CCC CCC CCC CCC
	GCA GCA CGT TAG ATC L350 GAG CTC	CAA GTT TAA ATT CCA CAC 400	260 GCC 1260 GCC CCC 1. CCC GCC CCC CCC CCC CCC CCC CCC CCC CC	AGA TOT 310 CCC GGG 13 AAA TTT	CAA GIT GCC CCC CCC GGG	CCT CGA ACC	ACC TCG  CGA GCT  ACC CGA GCT  CGA GCT  CGA GCT  CGA GCT  CGA GCT  CGA GCT	ACT TCA  TCG AGC  GCA CGT  370  ACT TCA	GGG CCC  1  GGC CCC  TCA AGT  ATA TAI  20  TGI	CAAC CAAC CTCC CACC CACC CACC CTCC	CAA CIT CAA 30 AAA TIT 1380 ATAT TAT	CCT CGA CCT CGA CCA AGC CCA CCGA CCGA CCGA CCGA	AAT TTA CCCC CCC 1 ACCC CCCC CCCC CCCC C	CTT CAA 340 CTC GAG 13 CAA CTC CTC	AAG TTC	CCA CCC CCC CCC CCC CCC CCC CCC CCC CCC
	GCA CGT TAG ATC	CAA GTT TAA ATT CCA GTG CAC	CGG GCCC 1.260 CCCC 1. CCCC GCCC CGCC CGCC CGCC CGCC C	AGA TCT 310 CCCC GGG	CAA GIT GCC GCC GCC 1410	CCT CGA ACT CTG CCC CCC CCC CCC CCC CCC CCC CCC C	ACC TCG CGA GCT AGG TCC CGA GCT CGA GCT CCC	ACT TCA ACC TCA ACC TCA ACC TCA ACC TCA ACC ACC	GGG CCCC  1  GGC CCCC  TCA  AGT  ATA  TAI  20  1470	CAA GTT	CAA CTT CAA 30 AAA TTT 1380 ATAT 1 CAC CTC	CCT CCA ACC AC	AAT TTA CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCC	CTT CAA 340 CTC CAC CAC CAC CAC CAC CAC CAC CAC CAC	GGA CCT TIT AAA AAG TTC AGG	CCA CCC CCC CCC CCC CCC CCC CCC CCC CCC

WO 96/40921 PCT/US96/09287

#### 26/41

### FIG. 5 D

1500 1510 1520 CTC ACC CTG TAG GTA TCT CAG TTC GGT GTA GGT CGT TCG CTC CAA GCT CAG TGC GAC ATC CAT AGA GTC AAG CCA CAT CCA GCA AGC GAG GTT CGA 1560 1570 1540 1550 GGG CTG TGT GCA CGA ACC CCC CGT TCA GCC CGA CCG CTG CGC CTT ATC CCC GAC ACA CGT GCT TGG GGG GCA AGT CGG GCT GGC GAC GCG GAA TAG 1590 1600 1610 1620 CGG TAA CTA TCG TCT TGA GTC CAA CCC GGT AAG ACA CGA CTT ATC GCC SCC ATT GAT AGC AGA ACT CAG GTT GGG CCA TTC TGT GCT GAA TAG CGG 1660 1650 1670 ACT GGC AGC AGC CAC TGG TAX CAG GAT TAG CAG AGC GAG GTA TGT AGG TGA CCG TCG TCG GTG ACC ATT GTC CTA ATC GTC TCG CTC CAT ACA TCC 1700 1710 1720 CGG TGC TAC AGA GTT CTT GAA GTG GTG GCC TAA CTA CGG CTA CAC TAG GCC ACG ATG TCT CAA GAA CTT CAC CAC CGG ATT GAT GCC GAT GTG ATC 1740 1750 1760 AMG GAC AGT ATT TOG TAT CTG CGC TCT GCT GAA GCC AGT TAC CTT CGG TTC CTG TCA TAA ACC ATA GAC GCG AGA CGA CTT CGG TCA ATG GAA GCC 1780 1790 1800 AAA AAG AGT TGG TAG CTC TTG ATC CGG CAA ACA AAC CAC CGC TGG TAG TIT TTC TCA ACC ATC GAG AAC TAG GCC GIT TGT TTG GTG GCG ACC ATC 1830 1840 1850 1860 1870 CCC TCC TIT TIT TCT TTC CAA CCA CCA GAT TAC CCC CAG AAA AAA ACC GCC ACC AAA AAA ACA AAC GIT CGT CGT CTA ATG CGC GTC TIT TIT TCC 1980 1890 1900 1910 ATC TCA AGA AGA TCC TTT GAT CTT TTC TAC GGG GTC TGA CGC TCA GTG THE ACT TOT ACE ANA CTA GAN AND ATE CCC CAG ACT GCG ACT CAC 1950 CAN CGA AAA CTC ACG TTA AGG GAT TTT GGT CAT GAG ATT ATC AAA AAG CTT GCT TIT GAG TGC AAT TCC CTA AAA CCA GTA CTC TAA TAG TIT TTC 1980 1990 2000 2010 CAT CTT CAC CTA GAT CCT TTT AAA TTA AAA ATG AAG TIT TAA ATC AAT CTA GAA GTG GAT CTA GGA AAA TIT AAT TIT TAC TTC AAA ATT TAG TTA 2020 2030 2040 2060 CTA AAG TAT ATA TGA GTA AAC TTG GTC TGA CAG TTA CCA ATG CTT AAT CAT TTC ATA TAT ACT CAT TTG AAC CAG ACT CTC AAT GGT TAC GAA TTA

#### **RECTIFIED SHEET (RULE 91)**

WO 96/40921 PCT/US96/09287

#### 27/41

### FIG. 5 E

2100 2070 2080 CAG TGA GGC ACC TAT CTC AGC GAT CTG TCT ATT TCG TTC ATC CAT AGT GTC ACT CCG TGG ATA GAG TCG CTA GAC AGA TAA AGC AAG TAG GTA TCA 2130 2120 2140 2150 2160 TGC CTG ACT CCC CGT CGT GTA GAT AAC TAC GAT ACG GGA GGG CTT ACC ACG GAC TGA GGG GCA GCA CAT CTA TTG ATG CTA TGC CCT CCC GAA TGG 2180 2190 2170 ATC TGG CCC CAG TGC TGC AAT GAT ACC GCG AGA CCC ACG CTC ACC GGC TAG ACC GGG GTC ACG ACG TTA CTA TGG CGC TCT GGG TGC GAG TGG CCC 2230 2240 TCC AGA TTT ATC AGC AAT AAA CCA GCC AGC CGG AAG GGC CGA GCG CAG AGG TCT AAA TAG TCG TTA TTT GGT CGG TCG GCC TTC CCG GCT CGC GTC 2280 2290 ANG TEG TEE THE AND TIT ATC CEC CTC CAT CCA GTC TAT THA TTG TTG TTC ACC AGG ACG TTG AAA TAG GCG GAG GTA GGT CAG ATA ATT AAC AAC 2310 2320 2330 CCG GGA AGC TAG AGT AAG TAG TTC GCC AGT TAA TAG TTT GCG CAA CGT GGC CCT TOG ATC TCA TTC ATC AAG CGG TCA ATT ATC AAA CGC GTT GCA 2360 2370 2380 2390 2400 TGT TGC CAT TGC TAC AGG CAT CGT GGT GTC AGG CTC GTC GTT TGG TAT ACA ACE GTA ACE ATG TCC GTA GCA CCA CAG TGC GAG CAG CAA ACC ATA 2410 2420 2430 2440 GGC TTC ATT CAG CTC CGG TTC CCA ACG ATC AAG GCG AGT TAC ATG ATC CCG AAG TAA GTC GAG GCC AAG GGT TGC TAG TTC CGC TCA ATG TAG TAG CCC CAT GTT GTG CAA AAA AGC GGT TAG CTC CTT CGG TCC TCC GAT CGT CCC CTA CAA CAC GTT TTT TCG CCA ATC GAG GAA GCC AGG AGG CTA GCA 2500 2510 2520 2530 TGT CAG AAG TAA GTT GGC CGC AGT GTT ATC ACT CAT GGT TAT GGC AGC ACA GTC TTC ATT CAA COG GOG TCA CAA TAG TGA GTA CCA ATA COG TCG 2550 2560 2570 2580 2590 ACT GCA TAX TTC TCT TAC TGT CAT GCC ATC CGT AAG ATG CTT TTC TGT TGA CGT ATT AAG AGA ATG ACA GTA CGG TAG GCA TTC TAC GAA AAG ACA 2600 2610 2620 2640 2630 CAC TGG TGA GTA CTC AAC CAA GTC ATT CTG AGA ATA GTG TAT GCG GCG CTG ACC ACT CAT GAG TTG GTT CAG TAA GAC TCT TAT CAC ATA CGC CGC

## FIG. 5 F

	650		26	560			2670			26	30		_	
	•			•			•				*			690
ACC GA	G TTG	CTC	TTG AAC	CCC	CCC	GTC CAG	AAC TTG	ACG TGC	CCT	TAA ATT	TAC ATG	ccc	<u>ссс</u>	ACA TGT
	2700			271	ro		27	720		:	2730			
TAG CA	G AAC C TTG	TTT AAA	AAA TTT	agt TCA	GCT CGA	CAT GTA	CAT GTA	TGG ACC	AAA TIT	ACC TCC	TTC AAG	TTC AAG	222 222	CCC
2740	2'	750		2	760			277	70		27	780		
AAA AC	T CTC	AAG TTC	GAT CTA	CTT GAA	acc TGG	GCT CGA	CTT CAA	CIC	ATC TAG	CAG GTC	TTC AAG	CTA	CTA CAT	ACC TGG
2790		280	00		28	310		2	2820			283	0	
CAC TO	C ACC	ACC TGG	CXX	CTG GAC	ATC TAG	TTC	AGC TCG	ATC TAG	TTT	TAC ATG	TTT XXX	CAC	CYC	CCT CCA
2840			2850			28	60 +		28	370		. 2	2880	
TTC TC	C CYC	AGC TCG	XXX TTT	AAC TTG	AGG TCC	TTC	CCT	XXX TTT	TGC ACG	CCC	λλλ ΤΤΤ	AAA TIT	CCC	AAT TTA
2	890		. 29	900.	-	. :	2910			292	20		2	930
AAG GC	C GAC	ACG TGC	CAA	ATG	TTG	AAT TTA	XCT TGX	CAT	ACT TGA	CIT	CCI	TTT	TCA	ATA
														444
	2940			29	50		29	960			2970			,
-	2940		_	29	•			960			2970			,
TTA T:	C AAG	CAT	TTA AAT	TCA	• GGG	TTA AAT	TIG	TCI	CAT GTA	GAG	œc	ATA TAT	CAT GTA	ATT
TTA TO AAT AA	C TTC	CAT	TTA AAT	TCA AGT	• GGG	TTA AAT	TIG	TCI	CIA	GAG	CCC	ATA TAT 020	CAT GTA	ATT TAA
AAT A	C AAG	CAT GTA 990 TTA	CAA	TCA AGT	GGG CCC 3000	ACA	TTG AAC	TCT AGA 30:	CTA 10 • CCT	CTC	3 600	TAT 020 CAC	GTA ATT	TAA
2980 TGA A	C AAG	CAT GTA 990 TTA	CAA	TCA AGT	GCC CCC 3000 TAA ATT	ACA	TTG AAC	TCT AGA 30: AGG TCC	CTA 10 • CCT	CTC	3 600	TAT 020 CAC	GTA ATT TAA	TAA
2980 TGA AT ACT TI	C AAG	CAT GTA 990 TTA AAT 30	GAA CTT	TCA ACT AAA TTT	GGG CCC 3000 TAA ATT	ACA TCT 050	ACA	TCT AGA 30: AGG TCC	GTA  10  GCT  CCA  3060	CAG CTC	CCC CCC 3 CCC CCC	CAC GTG 30	ATT TAA 70	TAA TCC AGG
2980 TGA AT ACT TI 3030	C AAG	CAT GTA 990 TTA AAT 30 GCC CGG	GAA CTT 40 ACC TGG	TCA AGT TTT TGA ACT	GGG CCC 3000 TAA ATT	ACA TGT 050 CTA GAT	AAT TTA	TCT AGA 30: AGG TCC	GTA  CCTA  GCTA  GCTA  CCAT  GTA	CAG CTC	GCG CCC TATT	CAC GTG 30 CAT GTA	ATT TAA 70 GAC CTG	TCC AGG
2980 TGA AT ACT TZ 3030 CCC AZ GGC TT	C AAG C TTC C TAT C ATA AGT T TCA	CAT GTA 990 TTA AAT 30 GCC CGG	GAA CTT 40 ACC TGG 3090	TCA AGT TTT TCA ACT	CCCC  TAA ATT  CCT CCA  TAT	ACA TGT 050 CTA GAT	AAT TTA AGA TCT	TCT AGA 30: AGG TCC AAC TTG	GTA  10  CCTA  3060  CAT  GTA  3	TAT ATA	CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCC	CAT	ATT TAA 70 GAC CTG	TCC AGG
2980 TGA ANACT TO 3030 CCG ANGCC TO 3086 AAC CTTTC GO	C AAG C TAT C ATA A AGT T TCA TATA A TAA A TAA	CAT GTA 990 TTA AAT 30 GCC CGG	GAA CIT 40 ACC TCG 3090 TAG ATC	TCA AGT AAA TTT TGA ACT	CCCC  TAA ATT  CCT CCA  TAT	ACA TGT 050 CTA GAT 31 CAC	AAT TTA AGA TCT	TOT AGA 30: AGG TCC AAC TTG	GTA  10  CCTA  3060  CAT  GTA  3	TATATATAC	CCCA	CAT	ATT TAA 70 GAC CTG 3120	TCC AGG
2980 TGA AT ACT TJ 3030 CCC AJ GGC TT 3080	C AAG C TAT C ATA AGT T TCA TA TAA AT ATT	CAT GTA 990 TTA AAT 30 GCC CGG	GAA CTT 40 ACC TGG 3090 TAG ATC	TCA AGT  TEA ACT  CCC CCC	GGG CCC 3000 TAA ATT 3 CCT GCA	ACA TGT 050 CTA GAT 31 CAC	AGA TCI OO CAG CTC 3150	TCT AGA 30 AGG TCC AAC TTG CCG	CTA CCA CAT GTA CTG CAC	TAT ATA ATG TAC	CCC CCC TATA ATA	CAT CTT	ATT TAA 70 CAC CTG 3120 TGC ACG	TAA TCC AGG ATT TAA GGC CCG
2980 TGA ANACT TO 3030 CCG ANGCC TO 3086 AAC CTTTC GO	C AAG C TAT C TAT C ATA AGT T TCA C ATA C	CAT GTA 990 TTA AAT 30 GCC CGG	CAA CTT 40 ACC TCG 3090 TAG ATC	TCA AGT TTT TCA ACT CCC CCC 140	GGG CCC 3000 TAA ATT 3 CCT GCA TAT ATA	ACA TGT 050 CTA GAT 31 CAC	AGA TCT  AGA TCT  GAG GCA GCCA GCCA GCCA	TCT AGA 30 AGG TCC AAC TTC	GTA  10  GCT  CCA  3060  CAT  GTA  3  CTG  GAC	TAT ATAC ACA ACA	CCC TAT ATA ATA ATA	CAC CTC 30 CAT CTA CTA	ATT TAA 70 CAC CTG	TAA TCC AGG ATT TAA
2980 TGA AT ACT TI 3030 CCG AI GGC TT 3086 AAC CT TTG GI	C AAG C TAT C TAT C ATA A AGT T TCA ATA ATA ATA ATA ATA ATA ATA ATA ATA	CAT GTA 990 TTA AAT 30 GCC CGG	GAA CTT 40 ACC TCG 3090 ATC	TCA AGT  TGA ACT  TGA ACT  TGT ACA	GGG CCC 3000 TAA ATT GCT GCA TAT ATA	ACA TOT  OSD  CTA GAT  CAC GTG	AGA ACT TTA ACA TCT CTC CTC CTC 3150	TCT AGA 30: AGG TCC AACC TTCC CGG CCGG	CTA CCA CAT CTA CTG CAC CAC CAC	TAT ATAT ATA ATA TATA	CCC CCC TATA ATA CCCA TCCCA TCCCA TCCCA TCCCA TCCCA TCCCA TCCCA TCCCA TCCCCA TCCCCCA TCCCCA TCCCA TCCCCA TCCCA TCCCCA TCCCCA TCCCCA TCC	CAC GTG GTA CTT GAA	ATT TAA  70 GAC CTG 3120 TGC ACG 3	TAA TCC AGG ATT TAA CCC CCG 170 AGA TCT
2980 TGA AT ACT TY 3030 CCC AI GGC TT 3086 AAC CC	C AAG C TTC  2 C TAT C ATA AGT T TCA AT AGT	CAT GTA 990 TTA AAT 30 GCC CCG	CAA CCIT 40 ACC TCG 3090 TAG ATC	TCA ACT  TGA ACT  GCG CCC  140  TGT ACA  31	GCC GCC TAA ATT GCA TAT ATA TCC ACG	ACA TGT 050 CTA GAT GAG CAC	AGA TOTA AGA TOT  GAG GCA GCA GCA GCA GCA GCA GCA GCA GC	TCT AGA 30: AGG TCC TTC CGG CCGG CCGG CCGG CCGG CCGG	GTA  10  GGT  CCA  3060  CAT  GTA  3  CTG  GAC	TAT ATA ATA TATA	CCC CCC TATA ATA CCCA ACCC TCCA TCCC TCCA TCCC	CAC GTG GTA CAT GTA CTT GAA	ATT TAA  70 GAC CTG 3120 TGC ACG TTCC	TAA TCC AGG ATT TAA CCG CCG

#### **RECTIFIED SHEET (RULE 91)**

## FIG. 5 G

3220		3:	230		:	3240			32	50		3:	260		
λλG	GAG	λCA	CTT	TAT	GTT	TAA	GAA	CCT	TGG	Τλλ	אַדער	رحت	ACC.	CCC	TTT AAA
3270			321	BO .		33	290		;	3300			33:	10	
CCC CCC	AGC TCG	CAA GTT	GCT CGA	AGA TCT	GAT CTA	CCC	GCT CGA	CTC CAC	GAA CTT	TGT ACA	GTG CAC	TCA AGT	GTT CAA	YCC TCC	GTG
3:	320		:	3330			334	10		33	350		3	360	
TGG ACC	AAA TTT	GTC CAG	CCC	AGG TCC	CTC GAG	CCC	age Teg	AGG TCC	CAG GTC	AAG TTC	TAT ATA	GCA CCT	AAG TTC	CAT.	GCA CCT
	331	70		33	380		:	3390			340	00		34	110
TCT AGA	CAA GTT	TTA AAT	CXC	AGC TCG	AAC TTG	CAG GTC	GCT CGA	CCC	CXG	CAG	GCA CGT	GAA CTT	GTA CAT	TGC ACG	AAA TTT
		3420				30		3	40			3450			
CCT CCT	TGC	ATC TAG	TCA AGT	ATT TAA	AGT TCA	CXG	CAA GTT	CCY	TAG ATC	TCC AGG	000 000	CCC	TAA ATT	CXC CXC	ဆ
3460			•			•				•			•		
CCX	TCC AGG	CCC	ccc	Τ <b>λλ</b> <b>λΤΤ</b>	CTC GAG	ecc 6CC	CCA	CAA CTT	CCC	CCC	λΤΤ Τ <b>λλ</b>	CIC	CCC	CCC	ATG TAC
3510			35	20		3:	530		:	3540			35	50	
CCI	GAC	Τλλ	TTT	TIT AAA	TTA	TTT	ATG	CAG	AGG	CCC	) CC	CCC:	<del>Сел</del>	*	CCI
CCY	GAC	Τλλ	TTT XXX	TIT	TTA AAT	TTT AAA	ATG TAC	CXC	agg TCC	222 223	AGG TCC	ecc ccc	CCT GGA	• CCC	CCJ CCJ
car cay	GAC CTG	TAA ATT	TTT	TIT AAA 3570 AGA	TTA AAT	TTT AAA AGT	ATG TAC 351	CAG GTC	AGG TCC	CCC CCC 3!	AGG TCC	CCC	CCT	CGG GCC	CCA
car cay	GAC CTG 560 AGC TCG	TAA ATT TAT ATA	TTT AAA TCC AGG	TTT AAA 1570 AGA TCT	TTA AAT	TTT AAA AGT	ATG TAC 351	CAG GTC	AGG TCC	CCC CCC 3!	AGG TCC	CCC	CCT	CGG GCC	CCA
GCT CGA 3! CTG GAC	GAC CTG 560 AGC TCG	TAX ATT TAT ATA	TTT AAA TCC AGG	TTT AAA 3570 AGA TCT	AGT TCA	ACT TCA	ATG TAC 351 GAG CTC	CIC CIC GIC GIC	AGG TCC	CCCC GGC 3! TIT AAA	AGG TCC 590 TTG AAC	CCC CCC	CCT GGA GCC CCG	CGG GCC 1600 TAG ATC	GCT CCA
GCT CGA 3! CTC GAC	GAC CTG S60 AGC TCG 361	TAT ATT ATA ATA	TTT AAA TCC AGG	TTT AAA S570 AGA TCT 30	AGT TCA	TTT AAA AGT TCA GGG	ATG TAC 351 CAG CTC	CAC GTC GTC GTC	AGG TCC GCT CGA	CCC CCC 3! TTT AAA	AGG TCC 590 TTG AAC	CCC GAC CTC	CCT GGA	CGG GCC 1600 TAG ATC	GCT CCA
GCT CGA 3! CTC GAC	GAC CTG 60 AGC TCG 361 GCA CCT	TAT ATT ATA ATA	TCC ACC	TTT AAA S570 AGA TCT 30	AGT TCA 520 CTT GAA	AGT TCA GGG CCC	ATG TAC 351 GAG CTC	CAG GTC GAG CTC 3630 ACC TCG	AGG TCC GCT CGA	CCG GGC 3! TIT AAA	AGG TCC 590 TTG AAC AGC TCG	CCC GGC GAG CTC	CCT GGA	CGG GCC 1600 TAG ATC	GCT CCA
GCT CGA 3! CTG GAC TTT AAA	GAC CTG 560 AGC TCG 361 GCA CCT	TAT ATA  TAT ATA  O  AAA  TTT  6660  CTC	TCC AGG	TITT AAA 1570 AGA TCT 30 TAG ATC	AGT TCA CTT GAA 367	AGT TCA CCC CCC	ATG TAC 351 GAG CTC	CAG GTC GAG CTC 3630 ACC TCG	AGG TCC	CAG GTC	AGG TCC 590 TTG AAC 364 AGC TCG	CCG GCC GAG CTC 10 ACC TGG	CCT GGA GCC CCG TTC AAG	CAC GTG	CCA CCA CCA CAT CTA
GCT CGA 3! CTG GAC TTT AAA	GAC CTG 560 AGC TCG 361 GCA CCT	TAA ATT TAT ATA TTT 6660 CTC GAG	TCC AGG	TITT AAA 1570 AGA TCT 30 TAG ATC	AGT TCA CTT GAA 367 TTC AAG	AGT TCA CCC CCC	ATG TAC 351 GAG CTC	CAG GTC GAG CTC 3630 ACC TCG	AGG TCC	CCC GGC 3: TIT AAA CAG GTC	AGG TCC 590 TTG AAC 364 AGC TCG	CCC GCC CAC CTC ACC TCG 3690	CCT GGA GCC CCC TTC AAG	CAC GTG	CCA CCA S50 CAT CTA
GCC CCC 3700	GAC CTG 560 AGC TCG 361 GCA CGT	TAA ATT  TAT ATA  O AAA TIT  G660 CTC GAG	TTT AAA TCC AGG AGC TCG AGC TCG	TIT AAA IS70 AGA TCT 36 ATC	AGT TCA CTT GAA 367 TTC AAG	AGT TCA  GGG CCC  O CCA GGT  1720	ATG TAC 351 GAG CTC CCC CCG	CAG GAG ACC TCG 36	GCT CCA	CCG GGC 3! TITT AAA CAG GTC	AGG TCC 590 TTG AAC TCG TCG	CCC GGC GAC CTC ACC TGG GFT CAA GTT	CCT GGA GCC CCG TTC AAG	CAC GTG	GCT CCA 550 CAT GTA CAT
GCT CGA  3! CTG GAC  TTT AAA  GCC CCG  3700	GAC CTG 560 AGC TCG 36: GCA CGT CAC GTG	TAA ATT TAT ATA TATA TATA TATA TATA TA	TTT AAA TCC AGG AGC TCG AGC TCG	TITT AAA IS70 AGA TCT 36 ATC	AGT TCA CTT GAA 367 TTC AAG	AGT TCA CCC CCC TCA CCCA TCA TCA	ATG TAC 351 GAG CTC CCC CCC CCC CCC	CAG GTC GAG ACC TCG GAA CTT	AGG TCC GCT CGA GCT CGA GTT CGA GTT CCA	CCG GGC 3! TITT AAAA CAG GTC	AGG TCC 590 TTG AAC 366 AGC TCG CAT GTA	CCC GCC CTC ACC TCC 3690 CAA GTT	CCT GGA GCC CCG TTC AAG	CAC GTG	GCT CCA 550 CAT GTA CAT
GCT CGA  3! CTG GAC  TTT AAA  GCC CCG  3700	GAC CTG 560 AGC TCG 36: GCA CGT CAC GTG	TAA ATT TAT ATA TATA TATA TATA TATA TA	TTT AAA TCC AGG AGC TCG AGC TCG	TITT AAA  1570 AGA TCT 36 TAG ATC  AAG TTC  CCA CGT	AGT TCA CTT GAA 367 TTC AAG	AGT TCA CCC CCC TCA CCT TCA ACT	ATG TAC 351 GAG CTC CCC CCC CCC CCC	CAG GTC GAG ACC TCG GAA CTT	GCT CCA GCT CCA GCT CCA GCT CCA GCT CCA GCT GCA GCT GCA GCT GCA GCT GCA GCT GCA	CCG GGC 3! TITT AAAA CAG GTC	AGG TCC 590 TTG AAC 366 AGC TCG CAT GTA	CCC GCC CTC ACC TCC 3690 CAA GTT	CCT GGA GCC CCG TTC AAG	CAC GTG	GCT CCA 550 CAT GTA CAT
GCT CGA  3! CTG GAC  TTT AAA  GCC CCG  3700  CTT GAA  3750  TGA	GAC CTG 560 AGC TCG 363 GCA CGT CAC GTG CAC	TAA ATT ATA ATA ATA ATA ATA ATA ATA ATA	TTT AAA TCC AGG AGC TCG AGC TCG TCG TCG TCG TCG	TITT AAA  1570 AGA TCT TAG ATC  AAG TTC  AAG ATC  AAG ATC	AGT TCA  367 CTT GAA  367 AGG	AGT TCA  GGG CCC  O CCA GGT  TGA ACT  ACT	ATG TAC	CAG GTC GAG GAG ACC TCG GAA CTT TCA	AGG TCC GCT CGA GCT GCA GTT CCA GCT CCA GCT CCA GCT CCA GCT CCA GCT CCA CCA CCA CCA CCA CCA CCA CCA CCA C	CCG GGC 3! TTT AAA CAG GTC AAA TTT AGC TCG	AGG TCC 590 TTC AAC AGC TCG CAT CTA CAT CTA	CCC GGC GGC CTC ACC TGG GTG GTA GTA CAT	CCT GGA GCC CCG TTC AAG GCA CGT TAT ATA	CAC GTG	CCA CCA CAT CTA CAT

## FIG. 5 H

								_								
	36	300		3	810			382	20		31	330		:	3840	
	TGA ACT	GCC.	CAA GTT	GTG CAC	TGT ACA	AGA TCT	AGA TCT	GTT CAA	ACC TGG	TGA ACT	GTG CAC	CIT	TTT <b>λλλ</b>	TGA ACT	TGG	CTC GAG
		385	50		38	360		:	3870			388	80		31	390
	TAG ATC	TAC ATG	CTT	TCA AGT	GTC CAG	TGA ACT	CCC	CTC	CAA GTT	CAG GTC	TGA ACT	CAT GTA	GTA CAT	TCT AGA	CAG GTC	222
		3	900			391	LO		35	20		3	930			
	TGT ACA	TGC	CAT	CTT CAA	TCG AGC	CCT	ccc	CTT GAA	ccc	CYC	AGA TCT	TCC AGG	CAA	CAA GTT	CCY CCY	CCY
394	0		39	50		3	960			397	0		39	80		
	CXX	CTG GAC	TGX ACT	YCY TCY	TTT AAA	CAA GTT	CTA CAT	CAA GTT	ccc	GAA CTT	œ6 6œ	TGC ACC	AGA TCT	GAC CTG	Caa Cit	TTT AAA
3	990			400	00		40	10		4	020			403	30	
	AAG TTC	CCI	CIC	CXC	TAA ATT	ACG TGC	GAT CTA	AAT TTA	CCT CCT	CAT GTA	CCY	CTC	CAA	CCA	CCY	CCC
	40	40		4	050			406	50		40	70		•	1080	
	CTG GAC	CTT CAA	TCC	AAT TTA	CCI	XCX TGT	GGA CCT	GTA CAT	TAC ATG	TCT AGA	CTA	CCC	AAC TTG	AGA TCT	TGG ACC	GCA CCT
															••	
		409	7 D		43	r00		4	1110			412	20		41	L30
	CCC	TTT	TGG	TTG	GCC	TTC	CAA GTT	TGG	CIT	TCC AGG	TGG	GCC	• CCA	AGG TCC	TCC	e GTA
	CCC	TTT	TGG	TTG AAC		TTC	GTT	TGG	CIT GAA	TCC AGG	TGG ACC	ecc soc	• CCA	AGG TCC	TCC	e GTA
	GGG TTA	TTT	TGG ACC	TGT	GCC	TTC AAG 41!	GTT 50 AGA	TGG ACC	CTT GAA 41 AGC	AGG L60 CTA	ACC	GCC GCC	CCA GGT 1170 GGA	TAT	TCC AGG	GTA CAT
416	GGG TTA	TTT	TGG ACC 1140 TGG ACC	TGT	GCC CCG	TTC AAG 41!	GTT 50 AGA	TGG ACC	CTT GAA 41 AGC	AGG L60 CTA	TGG ACC	GCC GCC	CCA GGT 1170 GGA CCT	TAT	TCC AGG	GTA CAT
416	TTA AAT	TTT AAA CTG GAC	TCG ACC 1140 TCG ACC 41	TGT ACA	222 222 223	TTC AAG 41! CGC GCG	GTT  50  AGA  TCT  1200  CTT	TCG ACC	CTT GAA 41 AGC TCG	AGG LEO CTA GAT 42:	TCG ACC	CAA	CCA GGT 1170 GGA CCT 4:	TAT ATA	TCC AGG	GTA CAT
	TTA AAT	TTT AAA CTG GAC	TCG ACC 1140 TCG ACC 41	TGT ACA	222	TTC AAG 41! CGC GCG	GTT 50 AGA TCT 1200 CTT GAA	TCG ACC	CTT GAA 41 AGC TCG	AGG LEO CTA GAT 42: TGG ACC	TCG ACC	CAA	CCA GGT 1170 GGA CCT 4:	TAT ATA	TCC AGG CCT GCA AGG TCC	GTA CAT
	TTA AAT  GGC CCG 1230	TTT AAA CTC GAC TCA AGT	TGG ACC 1140 TGG ACC 41 CTA GAT	TGT ACA	600 600 600 600 600 600	TITC AAG 41! CGC GCC GCC GCC GCC GCC GCC GCC GCC GC	GTT  50  AGA  TCT  1200  CTT  GAA  TCC	CAA GTA CAT	CTT GAA  41  AGC TCG  TGC ACG	AGG  CTA  GAT  42:  TGG  ACC	TGG ACC	CAA GTT	CCA GGT 1170 GGA CCT GAT CTA	TAT ATA 220 TAC ATG 42	AGG TCC	GTA CAT  GGA CCT  AAC TTG
	TTA AAT GGC CCG 1230 AAA TTT	TTT AAA CTC GAC TCA AGT	TGG ACC 1140 TGG ACC 41 CTA GAT	TGT ACA 190 CCG GGC 420	CXT	TITC AAG 41! CGC GCC GCC GCC GCC GCC GCC GCC GCC GC	GTT  50  AGA  TCT  1200  CTT  GAA  TCC	CAA GTA CAT	CTT GAA  41  AGC TCG  TGC ACG	AGG  CTA  GAT  42:  TGG  ACC	TGG ACC LO GGT CCA 1260 ACT	CAA GTT	CCA GGT 1170 GGA CCT GAT CTA	TAT ATA 220 TAC ATG 42 AGG TCC	AGG TCC	GTA CAT  GGA CCT  AAC TTG
	TTA AAT 10 CGC CCG 1230 AAA TTT 42	TTT AAA CTG GAC TCA AGT TGC ACG	TGG ACC 1140 TGG ACC 41 CTA GAT TGA ACT	TGT ACA 190 CCC GCC 420 CCA	CCC CCC CCC CCC CCC CCC CCC CCC CCC CC	TTC AAG 41! CSC CCC CCC CAC CCC CCC CAC CCC CCC CCC	GTT  1200 CTT GAA  TCC ACG	TCG ACC CAA GTT GTA CAT 250 CCA GGT 430	CTT GAA 41 AGC TCG TGC ACG TTGC TTGC TTGC TTGC TTGC TTG	AGG CTA GAT 42: TGG ACC CCT	TCG ACC LO GGT CCA ACT TGA CGT CGT	CAA CTT	CCA CCT CCT CTA CAT TTA	TAT ATA 220 TAC ATG 42 AGG TCC	TCC AGG CGT GCA AGG TCC TCG 4320	GTA CAT  GGA CCT  AAC TTG
	TTA AAT 10 CGC CCG 1230 AAA TTT 42	TTT AAA CTG GAC TCA AGT TGC ACG	TGG ACC L140 TGG ACC 41 CTA GAT TGA ACT AATT TTA	TGT ACA  190 CCC CCC  420 CCCA CCCA	CCC CCC CCC CCC CCC CCC CCC CCC CCC CC	TTC AAG 41! CSC CCC CCC CAC CCC CCC CAC CCC CCC CCC	GTT  1200 CTT GAA  TCC ACG	CAA GTA CAT CCA GGT 430	CTT GAA 41 AGC TCG TGC ACG TTGC TTGC TTGC TTGC TTGC TTG	AGG CTA GAT 42: TGG ACC CCT	TCG ACC LO GGT CCA ACT TGA CGT CGT	CAA CTT	CCA CCT 42 CAT TTA CCC CCC CCC	TAT ATA 220 TAC ATG 42 AGG TCC	TCC AGG CGT GCA AGG TCC TGG CAT GTA	CTA CCAT CCAT AAC TTC CTG GAC
	TTA AAT  GO CCC CCC AAA TTTT  42 TGA ACT	TTT AAA CTG GAC TCA AGT TGC ACG AGG TCC 43:	TGG ACC  1140  TGG ACC  41  CTA  GAT  TGA  ACT  AAT  ATTA	TOT ACA  190 CCC CCC  420 CCCA CCCA CCCA	CCC CCC CCC CCC CCC CCC CCC CCC CCC CC	TTTC AAG 411 CCC CCC CCC CCC CCC CCC CCC CCC CCC	AGA TCT GAA TCCT ACGA TCCT CTT TCCC ACGA TCCT TCCC TCCT TCCT	TGG ACC  CAA GIT  GTA CAT  CCA GGT  43  TCA AGT	AGC TCC TCC ACC CAC 4350	AGG CTA GAT 42: TGG ACC CCT CTG CAC	TGG ACC	CAA CTT CCA GGT GGT AAC	CCA CCT ATTA CCC GGC GGC GGC GGC GGC GGC GGC GGC GG	TCC TAT ATA 220 TAC ATG ACG TCC TTT AAA	TCC AGG  CGTA  AGG TCC  70  ACCC TGG  CAT  GTA  4.	GTA CAT  GGA CCT  AAC TTG  CTG GAC

## FIG. 5 I

		4380			43	90		4	400			4410			
CCC	CAT	TCC	TGG	GAA	CTG GAC	GAA CTT	TGG ACC	TGC	AGG	CIG	CCA GGT	TAC ATG	CAA	CTT	TAG
4420		4	430			4440			44	50		4	460		
GIG CAC	CAA	CCC	CAT	CCC	CCT	GGA CCT	CIT	TGG	TCT AGA	CIT	GCA CCT	CAT GTA	CCA	GGA CCT	CCC
4470			441	•			190			4500			453		
CAT GTA	CCI	CIT	ACT TGA	AAG TTC	CAA GTT	000 000	GCA CGT	CCC	GTA CAT	CCA	CAT	TCG	AGC	CIN	CCA CCT
	520			1530			454				550			560	GCT .
TCC	CAA	GGG	CCC	CCT	GGA	CAA	TGC	* CCG	TCC	TCT	GAC	TCC		•	~~~
λGG	GIT	ccc	ccc	GGA	CCI	CII	λCG	GGC	ACC	AGA	CIG	YCC	CYY	CCX	GCT -
	457	•			90			1590			460	1			510
TTG	CYC	CXX	CAT GTA	CXX	CCI	CII	TTC AAG	TGC ACG	TGG ACC	TGT ACA	CCC	CAA	TCG	CAG	TGC
		4620		• •	463				540			650			
CAG GTC	CAT GTA	CCC	CAT GTA	TCC AGG	000 000	CIC	TGT	CCC	CCA	CCI	CIT	CII	AGG TCC	TTA AAT	CIT
															-
4660			570			680			469				700		
TGA	AGA TCT	40 ccc	570 CCC	ccc	crc	TGC	CAA	· TTG:	469		ىنىن	47	700		
TGA	AGA TCT	40 ccc	570 CCC	ccc	crc	TGC 1680	CAA	· TTG:	465 TGA ACT	0	ىنىن	47	700	CIG	
TGA ACT 4710 AGC	CAT	CCC CCC	670 CCC GCC 472	CCC GGG	CTC GAG	TGC ACG	CAA GTT '30	TIG	465 TGA ACT	CCC CCC	CTT GAA	TGC ACG	AGT TCA 475	GAC CTG	AGA TCT
TGA ACT 4710 AGC TCG	CAT	CCC CCC	670 CCG GCC 472 CCG GGC	CCC GGG	CTC GAG	TGC ACG	CAA GTT '30	TTG AAC CAA CTT	465 TGA ACT	CCC GGG 1740 GAC CTG	CTT GAA	TGC ACG	AGT TCA 475 CGA GCT	GAC CTG	AGA TCT
TGA ACT 4710 AGC TCG 47	CAT GTA 760	CCT CCA	CCC GCC 472 CCC GGC	CCC GGG CAC GTG	CTC GAG ATC TAC	TGC ACG 47	CAA GTT 30 TCT AGA 478	CAA GTT	TGA ACT TGA ACT	CCC GGG 1740 GAC CTG	CTT GAA TCG ACC	TGC ACG	AGT TCA 475 CCA GCT	GAC CTG GCC CCG	AGA TCT CTT GAA
TGA ACT 4710 AGC TCG 47	CAT GTA 760	CCC CCC CCT CCA CCA CCA	CCC GCC 472 CCC GGC	CCC GGG 20 CAC GTG 1770 CTA GAT	CTC GAG ATC TAC	TGC ACG 47	CAA GTT 30 TCT AGA 478 CTT GAA	CAA GTT	TGA ACT TGA ACT	CCC GGG 1740 GAC CTG	CTT GAA TCG ACC	TGC ACC CCA CGT	AGT TCA 475 CCA GCT	GAC CTG 0 GCC CGG 800 TCC AGG	AGA TCT CTT GAA
TGA ACT 4710 AGC TCG CCA CCA TTC	CAT GTA 760 ATA TAT 481	CCC CCC CCA CCA CCA	CCC	CCC GGG	CTC GAG ATC TAC	TGC ACG 47 CCT GGA AGA	CAA GTT 730 TCT AGA 478 CTT GAA	CAA GTT TGA ACT	TGA ACT TGA ACT GTC CAC	CCC GGG 1740 GAC CTG 47 ATC TAG	TCG ACC	TGC ACE	AGT TCA 475 CCA GCT CTT GAA	GAC CTG O GCC CGG B00 TCC AGG	AGA TCT CTT GAA TAG ATC
TGA ACT 4710 AGC TCG CCA CCA TTC	CAT GTA 60 ATA TAT 481 ATC TAG	CCC CCC CCA CCA CCA	CCC	CCC GGG	CTC GAG ATC TAC	TGC ACG 47 CCT GGA ACA TCT	CAA GTT 730 TCT AGA 478 CTT GAA	TTG AAC CAA GIT TGA ACT 1830 ATC TAG	TGA ACT TGA ACT GTC CAC	CCC GGG 1740 GAC CTG 47 ATC TAG	TCG ACC	TGC ACE	AGT TCA 475 CCA GCT CTT GAA	GAC CTG O GCC CGG B00 TCC AGG	AGA TCT CTT GAA TAG ATC
TGA ACT 4710 AGC TGG TGG TTG ATT CCA AGT TTC AAG	CAT GTA 60 ATA TAT 481 ATC TAG	CCC CCC CCA CCA CCA CCA CCA CCA CCA CCA	CCC GCC	CCC GGG CAC GTG 1770 CTA GAT CGC GGG	ATT TAA ATT TAA CCCC GGG 487	TGC ACG ATT GGA TCT	CAA GTT 730 * TCT AGA 47E GAA GAG CTC	TTC AAC CAA GIT TCA ACT ACT TAG ATC TAG CTA	TCA ACT  GTC CAC  TTTT AAA	CCC GGG 1740 GAC CTG 47 ATC TAG	TCG ACC	TGC AGG CCA GGT AGC GGA CTT GAA CTT	AGT TCA 475 CCA GCT GAA CCT GCA	GAC CTG 60 * GCC CGG 8000 * TCC AGG 41 TAC	AGA TCT CTT GAA TAG ATC
TGA ACT 4710 AGC TGG TGG TTG ATT CCA AGT TTC AAG	CAT GTA 60 ATA TAT 481 ATC TAG	CCC CCC CCA CCA CCA CCA CCA CCA CCA CCA	CCC GCC	CCC GGG CAC GTG 1770 CTA GAT CGC GGG	ATT TAA ATT TAA CCC GGG 487 TTG AAC	TGC ACG ATT GGA TCT	CAA GTT 730 * TCT AGA 478 GAG CTT GAA TTT	TTC AAC CAA GIT TCA ACT ACT TAG ATC TAG CTA	TCA ACT  GTC CAC  TTTT AAA	CCC GGG 1740 GAC CTG ATC TAG	TCG ACC	TGC ACG CCA AGC CGT AGC TCG GAA CTT 890 ATT TAA	AGT TCA 475 CCA GCT GAA CCT GCA	GAC CTG 60 * GCC CGG 8000 * TCC AGG 41 TAC	AGA TCT CTT GAA TAG ATC

## FIG. 5 J

4950			49	60		49	970		•	4980			499	0	
CTA GAT	ATT TAA	GTT CAA	TGT ACA	GTA CAT	TTT AAA	TAG ATC	ATT TAA	CCA	ACC TGG	TAT ATA	GGA CCT	YCI TCY	GAT CTA	GAA CTT	TGG ACC
5	000		!	5010			50	20		5	030		:	5040	
GAG CTC	CAG GTC	TGG ACC	TGG ACC	AAT TTA	CCC	TTT AAA	λλΤ ΤΤλ	GAG CTC	GAA CTT	AAC TTG	CIG	TTT AAA	TGC ACG	TCA AGT	GAA CTT
	50	50		50	060		:	5070			500	30		50	90
CIT	ATG TAC	CCA GGT	TCT AGA	AGT TCA	GAT CTA	GAT CTA	CIC GYC	GCT CGA	ACT TGA	CCA	CIG	YCY YCY	CAA GTT	CAT GTA	TCT AGA
	:	5100			513	LO		51	L20 +		:	5130			
act TCA	CCI	CCA GGT	AAA TTT	AAG	AAG TTC	AGA TCT	AAG TTC	GTA CAT	CII	CIG	CCC	AAG TTC	GAC CTG	TTT AAA	CCI
5140			150 *			5160				•			<b>+</b>		
TCA AGT	CII	TTG AAC	CTA GAT	XGT TCX	TTT AAA	TTG AAC	AGT TCA	CXT GTX	GCT CGA	CAC	TTT AAA	) TCA	AAT TTA	AGA TCT	) TGX
5190			52	•			•			5220				•	
CYY	CCA	TCC	TTT AAA	GCT CGA	ATT TAA	TAC	<b>TGG</b>	<b>YCY</b>	AAG TTC	CIT	XXX TTT	CCA	CCI	CTG GAC	CTX GAT
53	240			5250			52	50		52	270			5280	
	•			•				•			•		_	•	
TAC	AAG TTC	aaa TTT	ATT	ATG TAC	Gλλ	λλλ	ТАТ	• TCT	GTA	λCC	TTT	λτλ	AGT	AGG	CAT GTA
TAC	TTC	AAA TTT	ATT	ATG TAC	CII CII	λλλ	TAT ATA	TCT AGA	GTA CAT	λCC	TTT AAA	λΤ <b>λ</b> Τ <b>λ</b> Τ	AGT	AGG TCC	CAT GTA
TAC ATG	525	TTT  O  TAT	<b>АТТ ТАА ААТ</b>	ATG TAC 53	CTT	AAA TTT	TAT ATA	TCT AGA 5310	CTA CAT	ACC TGG	TTT AAA 533	ATA TAT	AGT TCA	AGG TCC 5:	GTA 330
TAC ATG	525 AGT TCA	TTT  O  TAT	ATT TAA AAT TTA	ATG TAC 53	CTT	AAA TTT ATA TAT	TAT ATA CTG GAC	TCT AGA 5310 TTT AAA	CTA CAT TIT AAA	ACC TGG	TTT AAA 53: ACT TGA	ATA TAT	AGT TCA	AGG TCC 5:	GTA 330 CAT
TAC ATG	529 AGT TCA	TTT  OO  TAT  ATA  S340  TCT	ATT TAA AAT TTA	ATG TAC 53 CAT GTA	GAA CTT 100 AAC TTG 53!	AAA TTT ATA TAT	TAT ATA CTG GAC	TCT AGA 5310 TTT AAA 5:	GTA CAT TIT AAA	ACC TCG CTT GAA	TTT AAA 533 ACT TGA	ATA TAT  CCA GGT  S370	AGT TCA CAC GTG	AGG TCC 5: AGG TCC	GTA 330 CAT
AGA TCT	525 AGT TCA	TTT  OO  TAT  ATA  5340  TCT  AGA	ATT TAA AAT TTA GCT CCA	ATG TAC 53 CAT GTA ATT TAA	CAA CTT 100 AAC TTG 53! AAT TTA	AAA TTT ATA TAT	TAT ATA CTG GAC TAT ATA	TCT AGA 5310 TTT AAA 55	CTA CAT TIT AAA GGO CAA GTT	ACC TCG CTT GAA	TTT AAA 53: ACT TGA TTG AAC	ATA TAT  CCA GCT  S370 TCT ACA	AGT TCA CAC GTG	AGG TCC 5: AGG TCC	GTA 330 CAT GTA
AAC TTG  AGA TCT  5380	525 AGT TCA GTG CAC	TTT  OO  TAT  ATA  G340  TCT  AGA  5:	ATT TAA  AAT TTA  CCT CCA  390	ATG TAC 53 CAT GTA ATT TAA	GAA CTT 100 AAC TTG 53! AAT TTA	AAA TTT ATA TAT 50 AAC TTG	TAT ATA CTG GAC TAT ATA	TCT AGA 5310 TTT AAA 53 GCT CGA	CAT  TIT  AAA  GO  CAA  GIT  S4:	ACC TCC	TTT AAA 533 ACT TGA TTG AAC	ATA TAT  CCA GGT  S370 TGT ACA  ATG	ACC TGG	AGG TCC  S: AGG TCC  TTT AAA	GTA 330 CAT GTA
AAC TTG  AGA TCT  5380	525 AGT TCA GTG CAC	TTT  OO  TAT  ATA  G340  TCT  AGA  5:	ATT TAA  AAT TTA  CCT CCA  390	ATG TAC 53 CAT GTA ATT TAA	GAA CTT 100 AAC TTG 53! AAT TTA	AAA TTT ATA TAT 50 AAC TTG 5400 GTT CAA	TAT ATA CTG GAC TAT ATA	TCT AGA 5310 TTT AAA 53 GCT CGA	CTA CAT TIT AAA GTT S4: CAA CTT	ACC TCC	TTT AAA 533 ACT TGA TTG AAC	ATA TAT  CCA GGT  S370 TGT ACA  ATG	ACC TGG	AGG TCC 5: AGG TCC TTT AAA	CAT CAT CTA AGC TCG
AGA TCT 5380 TTT AAA 5430	SZS AGT TCA STC CAC TTA AAT	TTT  OO  TAT  ATA  GAO  TCT  AGA  SI  ATT  TAA	ATT TAA  AAT TTA  CCT CCA  TOT ACA  S4	ATG TAC SI CAT GTA ATT TAA AAA TTT CAT	GAA CTT BOO AAC TTG 533 AAT TTA	AAA TIT  ATA TAT  SO AAC TIG  SAIC TIG  CAA  CAC	TAT ATA CTG GAC TAT ATA ATA ATA CTAT ATA ATA CCA CCA	TCT AGA 5310 TTT AAA 5: CCT CCGA AAG TTC	CAA TITT AAA 160 CAA GIT CAA CAT CAA CAA	ACC TCG CTT GAA TTT 10 TAT ATA ATA ATT	TITT AAA  532 ACT TGA TTG AAC	ATA TAT  CCA GGT  5370  TCT ACA  ATC TAC	AGT TCA CAC GTG ACC TGG 420 TAT ATA 54°	AGG TCC 5: AGG TCC TTT AAA AGT TCA 70	CAT CAT CTA AGC TCG
AGA TCT 5380 TTT AAA 5430 TTG AAC	SZS AGT TCA STC CAC TTA AAT	TTT  OO  TAT  ATA  GAO  TCT  AGA  SI  ATT  TAA	ATT TAA  AAT TTA  CCT CCA  TOT ACA  S4  CAT CTA	ATG TAC SI CAT GTA ATT TAA AAA TTT CAT	GAA CTT BOO AAC TTG 533 AAT TTA	AAA TIT  ATA TAT  SO AAC TIG  SAIC TIG  CAA  CAC	TAT ATA CTG GAC TAT ATA ATA ATA CTAT ATA ATA CCA CCA	TCT AGA 5310 TTT ANA 5: GCT CGA AAG TTC TAC ATG	CAA TITT AAA 160 CAA GIT CAA CAT CAA CAA	ACC TCG CTT GAA TTT 10 TAT ATA ATA ATT TAA	TITT AAA  532 ACT TGA TTG AAC	ATA TAT  CCA GGT  5370  TCT ACA  ATC TAC	ACC TGG ACC TGG ACC TGG ACC TGG CCA	AGG TCC 5: AGG TCC TTT AAA AGT TCA 70	CTA  CAT CTA  AGC TCG  CCG CGG  ACT TGA
AAC TTG  AGA TCT  5380  TTT  AAA  5430  TTG  ACC  TTG  TTG  TTG  TTG  TTG  TTG	TTC  525 AGT TCA  GTG CAC  TTA AAT  ACT TCA  480 TTT	TAT ATA 5340 TCT AGA ATT TAA AGA TCT	ATT TAX  AAT TTA  ACT CCA  TOT ACA  CAT CTA	ATG TAC  SI CAT GTA  ATT TAA  AAA TTT  CAT GTA  CAT CCT	CAA CTT BOO AAC TTG S3! AAT TTA	AAA TIT  ATA TAT  SO AAC TTG  CAA CAG GTC  ACA	TAT ATA CTG GAC TAT ATA ATTA ATTA CCCA CCCT CCCT CCCT	TCT AGA 5310 TTT AAA 53 GCT CGA AAG TTC TAC ATG	CTA CAT TITT AAAA 360 CAA CTT CAC CTC CAC CTC	ACC TCG CTT GAA AAA TTT 10 TAT ATA ATA 5 GAA	TITT AAA  533 ACT TGA TTGA AAC  TTGT AAC  CCT  CCT	ATA TAT  20 CCA CGT TGT ACA ATG TAC  AGA TCT	ACC TGG ACC TGG ACC TGG ACA ACA	AGG TCC  SI AGG TCC  TITI AAA  AGT TCA  TTA  TTA  TTA  TTA  TTA	CTA  CAT CTA  AGC TCG  CCG CGG  ACT TGA

## FIG. 5 K

	55:	30		53	540		:	5550			556	50		51	570
		•			•			•				•			•
CIT	ACG	TTA	ACA	TGT ACA	ACA	ΤΑλ λΤΤ	CTT GAA	CAA	ATA	YCC YCC	AGC TCG	TTA AAT	TAA ATT	YCC	TTA AAT
	!	5580			559	90		56	500	•	9	610			
CAA GTT	ATA TAT	AAG TTC	CAA GTT	TAG ATC	CAT GTA	CAC GTG	λλλ TTT	TTT	CAC GTG	λλλ TTT	Τλλ λΤΤ	AGC TCG	<b>λΤΤ Τλλ</b>	TTT	TTC AAG
5620		56	530		9	640			56:	50		56	560		
ACT	GCA	TTC	TAG	TTG	TGG	TTT	GTC	CAA	λCT	CAT	CAA	TGT	ATC	тта	TCX:
TGA	CCI	AAG	ATC	AAC	ACC	λλλ	CAG	CII	TGA	GTA	CIT	λCλ	TAG	λλΤ	λGT
5670			568	30		56	590		:	5700			571	r0	
TGT ACA	CIC CIC	GAT CTA	CTC GAG	TAG ATC	CTT GAA	CCT GCA	CXC CXG	AAG TTC	CIG GXC	GGT CCA	GAC CTG	TGC ACG	agt TCA	GAA CTT	TAA ATT
57	720		:	5730			574	10		57	750		:	5760	
TAA ATT	λλΤ ΤΤλ	CXC	TGT ACA	TTG AAC	TCC AGG	GAA CTT	<b>λΤλ ΤλΤ</b>	ccc	GTT CAA	TTG AAC	AGA TOT	TTT AAA	CIG	TCG AGC	CCC
	57	7D		57	780		:	5790			580	00		51	B10
. ACT TGA	λλλ TTT	TTC	ATG TAC	TCG AGC	CCC	CTX	agt TCA	CCA	CXX CXX	TAT ATA	ccc ccc	CCX	TAG ATC	AGA TCT	TGG
	!	5820			5B:	30		5	340		:	5850			
CGA		•	λλλ	AAT		•	TTG		•	TGG		•	GAA	እእጥ	CTYC*
GCT	ТАТ	TGG	AAA TTT	AAT TTA	CGA	• TAT	TTG AAC	λλλ	λτλ	TGG ACC	CAT	ATT	GAA CIT	AAT TIA	CYC
CGA GCT 5860	ТАТ	TGG	AAA TTT 370	AAT TTA	CGA GCT	• TAT	TTG	λλλ	λτλ	ACC	CAT	ATT TAA	CAA CTT	AAT TIA	GTC CAG
5860 • CCC	TAT ATA	TGG ACC 51	TTT 370 AGT	AAT TTA TTC AAG	CCA GCT	TAT ATA 5BB0	AAC	AAA TIT	ATA TAT 589	ACC 90 • GCC	CAT GTA	ATT TAA 59	CIT CIT	TTA	CAG
5860 • CCC	TAT ATA	TGG ACC 51	TTT 370 AGT	TTA	CCA GCT	TAT ATA 5880 GTA CAT	AAC	AAA TIT	ATA TAT 589 ATC TAG	ACC 90 • GCC	CAT GTA	ATT TAA 59	CIT CIT	AAA TTT	CAG
5860 GCC CCC 5910	TAT ATA GAT CTA	TGG ACC 51 GTG CAC	TTT  370  AGT TCA  59:	TTA TTC AAG	CCA GCT TCT ACA	TAT ATA 5880 GTA CAT 59	ACT TGA 330 CTG	AAA TITI CAT CTA	ATA TAT 589 ATC TAG	ACC GCC GCC GCG S940	CAT GTA ATT TAA	ATT TAA 55 TTT AAA	CTT 900 CCA GGT 59:	AAA TIT	CAG CTC CAC
5860 GCC CCC 5910	TAT ATA GAT CTA	TGG ACC 51 GTG CAC	TTT  370  AGT TCA  59:	TTA TTC AAG	CCA GCT TCT ACA	TAT ATA 5880 GTA CAT 59	ACT TGA 330 CTG	AAA TITI CAT CTA	ATA TAT 589 ATC TAG	ACC GCC GCC GCG S940	CAT GTA ATT TAA	ATT TAA 55 TTT AAA	CTT 900 CCA GGT 59:	AAA TIT	CAG CTC CAC
SCT 5860 CCC CCC 5910 ATT TAA	TAT ATA CAT CTA TTT AAA	TGG ACC SI	ACT TCA 592 CAT GTA	TTA TTC AAG ACG TGC	CCA CCT TCT ACA CCA CCT	TAT ATA 5880 GTA CAT 59	ACT TGA 930 CTG GAC	CAT CTA	ATA TAT 589 ATC TAG ATA TAT	ACC  GCC CCC  S940  GCC CCC  SCC  SSS	CAT GTA ATT TAA CTT GAA	ATT TAA 59 TTT AAA ATA TAT	CTT  900  CCA  GGT  59:  TCG  AGC	AAA TTT 50 TTT AAA	CAG CAC CAC
GCT 5860 GCC CCC S910 ATT TAA S:	CAT CTA	TGG ACC 51 GTG CAC	ACT TCA 592 CAT GTA GAT	TTA TTC AAG 20 ACG TGC 5970	CCA GCT TCT ACA CCA GCT	TAT ATA 6880 GTA CAT 5! TAT ATA	ACT TGA 930 CTG GAC 599	CAT CTA CCC CCC	ATA TAT 589 ATC TAG ATA TAT	ACC  GCC CCC  S940  GCC CCC  S5940  GCC CCC  S59	CAT GTA ATT TAA CTT GAA	ATT TAA  55 TTT AAA  ATA TAT	CTT  900  CCA  CCT  59:  TCG  ACC	AAA TTT AAA 6000	CAG CTC CAC
GCT 5860 GCC CCC S910 ATT TAA S:	CAT CTA	TGG ACC 51 GTG CAC	ACT TCA 592 CAT GTA GAT	TTA TTC AAG 20 ACG TGC 5970 AGA TCT	CCA GCT TCT ACA CCA GCT	TAT ATA 6880 GTA CAT 5! TAT ATA	ACT TGA 330 CTG GAC 591 TGG ACC	CAT CTA CCC CCC	ATA TAT 589 ATC TAG ATA TAT	ACC  GCC CCC  S940  GCC CCC  S5940  GCC CCC  S59	CAT GTA ATT TAA CTT GAA	ATT TAA  59 TTT AAA  ATA TAT  TTC AAG	CTT  900  CCA  CCT  59:  TCG  ACC	AAA TTT 50 TTT AAA 6000	CAG CTC CAC ACG TCC
SECT SB60 CCC S910 ATT TAA SCCC	CAT CTA TTT AAA 960 CAT CTA AAA	TGG ACC STG CAC CAC CAC CCC CCC CCC CCC CCC CCC CC	TTT  370  AGT TCA  593  CAT GTA  GAT CTA	TTA TTC AAG 20 ACG TCC 5970 AGA TCT	CGA GCT TCT ACA CGA GCT	TAT ATA  GTA CAT  TAT ATA  CIT GAA	ACT TGA 330 CTG GAC 599 TGG ACC	CAT CTA CCTA CCCC CCC TCA ACT TCA TCA	ATA TAT 589 ATC TAG ATA TAT CITT GAA	ACC	CAT CTA ATT TAA CTT CAA 990 CCA CCT 60	ATT TAA 5: TITT AAA ATA TAT TAC AAG	CTT  900  CCA  GGT  599  TCG  AGC	AAA TIT  50 TIT AAA 6000 CIC	CAG GTG CAC ACG TGC TGC
SECT SB60 CCC S910 ATT TAA SCCC	CAT CTA TTT AAA 960 CAT CTA ATA TAT	TGG ACC STG CAC CAC CAC CCC CCC CCC CCC CCC CCC CC	TTT  370  AGT TCA  593  CAT GTA  GAT CTA	TTA TTC AAG 20 ACG TCC 5970 AGA TCT	CGA GCT TCT ACA CGA GCT CGA GCT CGA CCGA C	TAT ATA  SERIE CAT  SETAT ATA  CIT GAA  TAT ATA	ACT TGA 330 CTG GAC 599 TGG ACC	CAT CTA CCTA CCCC CCC TCA ACT TCA ACT	ATA TAT 589 ATC TAG ATA TAT CITT GAA	ACC	CAT CTA ATT TAA CTT CGAA 990 CGA CCT 60 ATA TAT	ATT TAA 5: TITT AAA ATA TAT TAC AAG	CCA S910 CCA SGT TCC AGC TGT ACA CCC	AAA TIT  50 TIT AAA 6000 CIC	CAG CAC ACG TGC ACG ACC ACC
SCT S860 CCC S910 ATT TAA SS CCC CCC	CAT TITT AAA  CTA TITT AAA  GAT CTA  CTA  CTA  CAT CTA  CAT CTA  CAT CTA  CAT	TGG ACC STG CAC CCC CCC CCC CCC CCC CCC ACC ACC AC	TITI  AGT TCA  599 CAT GTA  CAG CTA  CAG CTA  CAG CTA  CAG CTA	TTA TTC AAG 20 ACG TGC 5970 AGA TCT TTT AAA	CGA GCT TCT ACA CGCT CCCA CCCT CCCA CCCT ATCC ATCC	TAT ATA  SBB0  CTA CAT  SS  TAT ATA  ATA  TAT ATA	AACI TGA 330 CTG GAC 599 TGG ACC	AAA TITT CAT CCTA CCCC CCC TCA ACT TCA ACT 6	ATA TAT  589 ATC TAG ATA TAT  CTIT GAA  CAG GTC	ACC SCC CCC SS ACC CCC CCC CCC CCC CCC C	CAT CTTA ATT TAA CTT CGAA 990 CGA CCT 600 ATTAT	ATT TAA SE TITT AAAA ATA TAT TACT AAG 6090	CTT 900 CCA CCT 599 TCG AGC TGT ACA ACA	AAA TIT 50 TIT AAA 60000 GIC CAC TAT ATA	CAG CTC CAC ACG TGC TGC ACG ATC ATC
SCT S860 CCC S910 ATT TAA SS CCC CCC	CAT TITT AAA  CTA TITT AAA  GAT CTA  CTA  CTA  CAT CTA  CAT CTA  CAT CTA  CAT	TGG ACC STG CAC CCC CCC CCC CCC CCC CCC ACC ACC AC	TITI  AGT TCA  599 CAT GTA  CAG CTA  CAG CTA  CAG CTA  CAG CTA	TTA TTC AAG 20 ACG TGC 5970 AGA TCT AAA	CGA GCT TCT ACA CGCT CGA GCT ACA TTAG	TAT ATA  SBB0  CTA CAT  SS  TAT ATA  ATA  TAT ATA	AACT TGA 330 CTG GAC 599 TGG TCC AGG	AAA TITT CAT CCTA CCCA CCCC TCA ACT 6030 ACT ACT 60CA	ATA TAT  589 ATC TAG ATA TAT  CIT GAA  CAG GTC	ACC CCC CCC CCC CCC CCC CCC CCC CCC CCC	CAT TAA CIT GAA CCT GO ATAT TAT CAA CTT	ATT TAA SE TITT AAAA ATA TAT TACT AAG 6090	CTT 900 CCA CCT 599 TCG AGC TGT ACA ACA	AAA TIT 50 TIT AAA 60000 GIC CAC TAT ATA	CAG CAC ACG TGC ACG ACC ACC

## FIG. 5 L

6100		6:	110		1	6120			61	30		6	140		
TAT ATA	aca TGT	TTG AAC	AAT TTA	CAA GTT	TAT ATA	TGG ACC	CCA	ΤΤ <b>λ</b> λλΤ	GCC CCC	ATA TAT	TTA AAT	TTC AAG	ATT TAA	CCX	Τ <b>λ</b> Τ <b>λ</b> Τλ
6150 +			61	60 •		6	170		(	6180			61	90	
λΤ <b>λ</b> ΤλΤ	GCA CCT	Τλλ ΑΤΤ	λTC TλG	AAT TTA	λΤΤ Τλλ	CCC	TAT ATA	TGG	CCA GGT	Jal. 2	CAT	ACG TCC	TTG	TAT	CCA
	00			5210				20			230			71A 5240	GGT
TAT	CAT	AAT	ATG	TAC	ATT	TAT	ATT	• GGC	TCA	TGT	ccy •	ХСХ		*	ccy.
ALA (	625		TAC		TAA 260	ATA		270		λCλ			AAT		CCY
		•			*			•			628	•			190
ACA	TGA ACT	CAT	TGA ACT	TTA AAT	TTG AAC	XCT TGX	AGT TCA	TAT ATA	TAA ATT	TAG ATC	Τλλ λ <b>1</b> Τ	TCA AGT	ATT TAA	ACC TCC	CCC
		300				•			•			330			
TCA (	TTA AAT	CAA	CAT GTA	AGC TCG	CCX	TAT ATA	ATG TAC	CIC	TTC AAG	<u>cc</u> cc	CYY	ACA TGT	Τλλ λΤΤ	CYY CILI	ACG TGC
5340 •			50	•		•				•			80		
CAT	AAT TTA	CCC	ccc	CCI	CCC	TCA ACT	ccc	CCC	AAC TTG	CIC	ecc ccc	ecc 0	CCY	TTG AAC	ACG TGC
6390			640	00		64	110		6	420			643	0	
TCA A	XTX TXT	ATG TAC	YCC TCC	TAT ATA	CYY	CCC	ATA TAT	CTA CAT	acc TCC	CCY CCY	<b>λΤλ</b> <b>ΤλΤ</b>	CCC CCC	ACT TGA	TTC AAG	CAT GTA
64	40		•	450		•	646	0		64	170		•	480	
	•			•				•			•				
TGA (	CCT	CLI	TCC	CXC	CTC	TAT ATA	TTX AAT	CCC	TAA	<b>TGX</b>	CCC	CAC	AYC YYC	CCI	CTX CXT
	649	0		65	500		•	5510			652	20		69	530
CAT (	CAA	CYC	TAT ATA	CAT GTA	ATG TAC	CCX CCT	AGT TCA	ACG TGC	CCC	CCT GGA	<b>λΤΤ Τλλ</b>	CIG	CTC CAG	λλΤ ΤΤλ	GAC CTG
	6	540			655	50		6	560	•	(	6570			
GGT .	λλλ	TCC	ccc	GCC	TGG	CAT	ТАТ	GCC	CXG	TAC	λTG	ACC	TTA	TCC	GAC
CCX	TTT	YCC	CCC	CCC	ACC	CTA	ATA	CCC	CIC	λTG	TAC	TGG	AAT	ACC	CIC
6580		6:	90		•	5600			66	10		6	620		
TTT	CCT	ACT	TCC	CXC	TAC	ATC	TAC	GT3	TTA	<u>.</u>	ልጥሮ	CCT	יידע *	ACC.	ATG
222	CCY	TGA	YCC	GTC	ATG.	TAG	ATG	CAT	λλΤ	CAG	TAG	CCA	TAA	TCG	TAC
6630			66	40		6	650			6660			66	70	
CYC	ATG TAC	CCC CCC	TTT	TCC	CAG	TAC	ATC TAG	AAT TTA	CCC	CCA	GGA	TAG	CCC	TIT	CIG

## FIG. 5 M

6	680			6690			67	00		6	710			6720	
TCA AGT	CCC	CCI	TTT AAA	CCA	AGT TCA	CTC GAG	CAC GTG	CCC	ATT TAA	GAC CTG	CAG	AAT TTA	CCC	AGT TCA	TTG AAC
	67	30		6	740			6750 *			67	60		6	770
TTT	TGG	CAC	CAA GTT	AAT TTA	CAA GTT	CCC	GAC CTG	TTT AAA	CCA GGT	AAA TTT	TGT ACA	CGT	AAC	AAC TTG	TCC AGG
		6780			679	90		61	900		•	5810		•	
939 900	CCA	TTG AAC	ACG TGC	CAA GTT	ATG TAC	CCC	CCA CCA	AGG TCC	CCT CCA	GTA CAT	CCC	TGG	CIC	GTC CAG	TAT ATA
6820 *			330			5840			685	*			960		٠
ATA TAT	AGC	AGA TCT	CCA	CCA	TTX AAT	CAC	AAC	CCI	CLC CYC	ATC TAG	CCC	TCC	AGA TCT	000 000	CAT
6870 •			688	•		-	390			900			691	•	
CCY	ccc	TCT ACA	TTT AAA	GAC	CIC	CTA	AGA TCT	YCY TCT	CAC	ccc	CTC	cci.	TCC AGG	AGC TCG	CTC
	920			930			694	•	•		50			960	
223 223	CCC	ecc ecc	CII	CCC	YCC YCC	XTT TXX	CCI	YCC TCC	CCC	ATT TAA	CCC	CCY	ccc ccc	AAG TTC	) TCA
	697	70		69	980			5990		•	700	00		70	)10
		•			•			•				•			
CIG	GTA CAT	AGT TCX	ACC TGG	000 000	TAT ATA	AGA TCT	GTC CAG	TAT ATA	λGG	ccc	ACC	•	TTG AAC	CCT	*
CIG	CAT 7	7CA 7020	TGG	ಯಾ	703	O ICI	CAG	70	AGG TCC	ccc	ACC TGG	050	AAC	ocy eci	TCT AGA
TAT	CAT 7	TCA 020 TGC	TAT	೦೦೦	703 GTT	TTT	CAC	ATA 70	AGG TCC	222 222 227	ACC TGG 7	050	AAC	GCT CCA	TCT AGA
TAT	CAT 7	TCA 020 TGC ACG	TAT	ACT	703 GTT CAA	TTT	ccc ccc	ATA 70	AGG TCC	CCC	ACC TGG 7	CCC GCG CCG CAC GTG	AAC	GCT CCA	TCT AGA
TAT ATA 7060	CAT CCA CCT ATC	TCA  O20  TGC ACC  70  TTA	TAT ATA	ACT TGA	703 GTT CAA	TCT 10 TTT AAA 7080	CAG	70 TTG AAC	AGG TCC 040 GGG CCC 709	CCC GGG	ACC TGG 7 ATA TAT	CCC GGG 7050 CAC GTG	CCC GGG	888 888	TCT AGA TTC AAG
TAT ATA 7060	CAT CCA CCT ATC	TCA  O20  TGC ACC  70  TTA	TAT ATA	ACT TCA CTC	703 GTT CAA	TCT TTT AAA OBO CTA CAT	CAG	70 TTG AAC	AGG TCC 040 GGG CCC 709 AGC TCG	CCC GGG	ACC TGG 7 ATA TAT	CCC GGG 7050 CAC GTG	CCC GGG	CCT CCA CCC CCC	TCT AGA
7060 CTC GAG 7110	CAT CCAT ATC TAC CAT	TCA TCC ACC TTA AAT TAT	TAT ATA TAG ATC 712 TGA	ACT TGA CTC CAC	TAC	TCT  TTT  AAA  OBO  CTA CAT  71	CAG CGC CCG TAG ATC	TTG AAC	AGC TCC 709 AGC TCG	TCT AGA	ACC TGG 7 ATA TAT TAG ATC	CCC GCG CAC GTG 71 GTG CAC	CCC GGG	CCT CCA CCC CCA CTT	TCT AGA TTC AAG ATT TAA
TAT ATA 7060 CTC GAG 7110 GAC CTG	CAT CCAT ATC TAC CAT	TCA TCC ACC TTA AAT TAT	TAT ATA 70 TAG ATC 712 TGA ACT	ACT TCA CTC CAC	TAC	TCT  TTT  AAA  OBO  CTA CAT  71	CAG CGC CCG TAG ATC	TTG AAC CIT GAA	AGC TCC 709 AGC TCG	TCT AGA CTA GAT	ACC TGG 7 ATA TAT TAG ATC	CCC GCG CAC GTG 71 GTG CAC	CCC GGG  TGG ACC  715  CCA GGT	CCT CCA CCC CCA CTT	TCT AGA TTC AAG ATT TAA
TAT ATA 7060 CTC GAG 7110 GAC CTG	CAT CCT ATC TAC CAT CTA CAT	TCA TCC ACC TTA AAT TAT ATA	TAT ATA 70 TAG ATC 712 TGA ACT	ACT TCA CTC CAC CCA CCT 170	TAC  CTC CAA  CTC CAC  CTC CAC	TCT TTT AAA TOBO GTA CAT TCC GGG	CAG GGC CCG TAG ATC TAT ATA T1E	TTG AAC	AGG TCC  140  GGG CCC  709  AGC TCG  TGA ACT	CCC GGG  TCT AGA  CTA GAT  CGA GCT  71	ACC TGG 7 ATA TAT TAG ATC TAC ATG	OSO	CCC GGG  TGG ACC  715  CCA GGT	GCT CCA CCC GCC GCC GTT CAA TTA AAT	TCT AGA TTC AAG ATT TAA CTA GAT
TAT ATA 7060 CTC GAG 7110 GAC CTG	CAT CCT ATC TAC CAT CTA CAT	TCA TCC ACC TTA AAT TAT ATA ATA	TAT ATA 70 TAG ATC 712 TGA ACT	ACT TCA CTC CAC	TAC  CTC CAA  CTC CAC  CTC CAC	TCT TTT AAA TOBO GTA CAT TCC GGG	CAG GGC CCG TAG ATC TAT ATA TIE CAC GTC	TTG AAC	AGG TCC  140  GGG CCC  709  AGC TCG  TGA ACT	CCC GGG  TCT AGA  CTA GAT  CGA GCT  71	ACC TGG 7 ATA TAT TAG ATC TAC ATG	OSO CAC TITT AAA	CCC GGG  TGG ACC  715  CCA GGT	GCT CCA CCC GCC GCC GCC TTTA AAT 2000 TAT ATA	TCT AGA TTC AAG ATT TAA CTA GAT
TAT ATA 7060 CTC GAG 7110 GAC CTG TAG	CAT CCAT ATG TAC CAT GTA 60 CAT GTA 721 ACA	TCA TCC ACC TTA AAT TAT ATA ATC TTC	TAT ATA TAG ATC TICA ACT TAG ACT TAG TAC TCC	ACT TCA CTC CAC	703 GTT CAA TAC ATG TAC CTC GAG CTT GAA	TCT TTT AAA TOSO CTA TTC CCC CCC CCC CCC CCC CCC CCC CCC	CAG GGC TAG ATC TAT TAT TEA TEA	TTG AAC  CIT GAA  TGG ACC  ACC  CIT CAA  COAC  C	AGG TCC  40  40  CCC  709  AGC TCG  TGA ACT  TCT AGA	TCT AGA  CTA CGAT  CTT CGAA	ACC TGG 7 ATA TAT TAG ATC TAC ATG 90 TAT ATA 724	TTTT AAAA	AAC CCC GGG OO TCG ACC 715 CCA GGT TTA	GCT CCA CCC GCC GCC GCC TTA AAT TAT ATA	TCT AGA TTC AAG ATT TAA CTA GAT GCC CCG

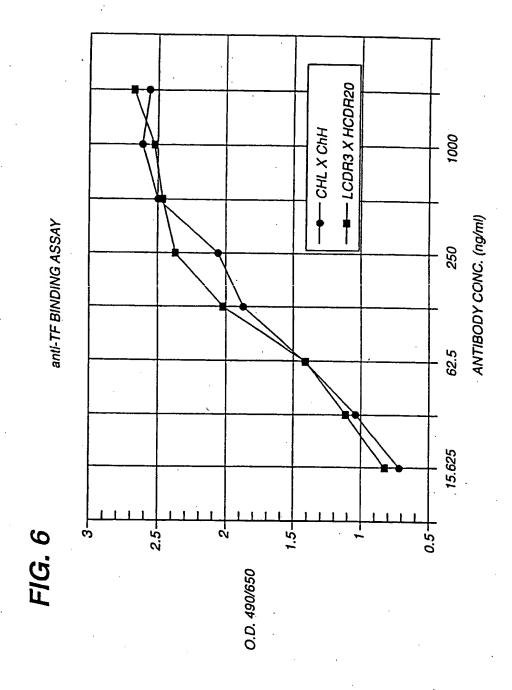
## FIG. 5 N

	7260				70			280			7290			
TGG GGT ACC CCA	CIC CAG	TAA	TAT ATA	TAT ATA	TTA AAT	CAA GTT	ATT TAA	CAC	ATA TAT	TAC	AAC	ACC TGG	ACC	GTC
7300 +	_	310			7320				30			340		
CCC AGT GGG TCA	CCC	000 000	AGT TCA	TTT AAA	TAT	TAA ATT	ACA TGT	TAA ATT	CCT	ccc	ATC	TCC	λCG	CGA
7350		73				370			7380		170	73:		GCT
300 000			•			. •			•					
ATC TCG TAG AGC	CCA	TGC	ACA	AGG	CCI	CIX	ccc	CIC	TTC	TCC AGG	CCY	YCC TCC	CCC	CCI
7400			7410			742				130			7440	
GCT TCT CGA AGA	λCλ	TCC	GAG	ccc	ייבאני	-	CD M		-	•			•	
		λGG	CIC	GGG	λCG	λGG	CIN	œ	AGG	TOG	CIC	TCA AGT	TCC	TCG
74!	•						7470			748	_			190
CLC CCC	AGC	TCC	TTC	CIC	CIA	<b>ACA</b>	CTC	GAG	CCC	λGλ	CIT	λGG	CAC	ACC
		AGG	XXC	بس.	CA.I	TCT	تبب	CIC	CCG	TCT	CAA	TCC	CIC	TCG
•	7500			751	.0		75	20		7	7530	,		
ACG ATG TGC TAC	CCC	ACC TGG	ACC TGG	ACC TGG	AGT	GTG	œ	CAC	λλG	GCC	CIC.	ထေ	GTA	CCC
						~~~	~~		TIL	122		$\sim$	^	
7540		50			560					CCC			CAT	ccc
7540 •	75	50		7	560			757	10		75	80		
7540 • TAT GTG	75 TCT	50 •	AAT	7 GNG	560	~~~		757	70		75	80		
7540 •	75 TCT	50 •	AAT TTA	7 GNG	CTC CAG	~~~		757 CCC GCC	70		75	80	CAC CTG	
7540  TAT GTG ATA CAC  7590  TTT GGA	75 TCT AGA	SAA CTT 760	AAT TTA 0	CAC CTC	S 60 CTC CAG 76 CCG	CCC	CYC CTC	757 CCG GCC 7 GAA	CCT CCA 620	TGC ACG	75 ACC TCG	GCT CCA 763	CAC CTG 0	GCA CGT
7540  TAT GTG ATA CAC  7590  TTT GGA AAA CCT	75 TCT AGA	SAA CTT 760	AAT TTA 0	CAC CTC	S 60 CTC GAG 76	CCC	CYC CTC	757 CCG GCC 7 GAA	CCT CCA 620	TGC ACG	75 ACC TCG	GCT CCA 763	CAC CTG 0	GCA CGT
7540 * TAT GTG ATA CAC 7590 * TTT GGA AAA CCT 7640	75 TCT AGA AGA TCT	50 CAA CTT 760 CTT CAA	AAT TTA 0 • AAG TTC	CIC CIC CCC CCA CCI	76 CCC CCC CCC	600 10 600 600 600 766	CAC CTC	757 CCG GCC 7 GAA CTT	GCT CCA 620 GAT CTA	TGC ACG GCA CGT	75 ACC TCG	GCT CGA 763 AGC TCG	GAC CTG 0 • TGA ACT	CCY CCI CCX
7540  TAT GTG ATA CAC  7590  TTT GGA AAA CCT	75 TCT AGA AGA TCT	50 GAA CTT 760 CTT GAA 7	AAT TTA 0 AAG TTC 650	CIC CIC CCIC	560 CTC GAG 76 GCG CCC	CCC CCC 10 CCA CCT 766	CAC CTC	757 CCC GCC 7 GAA CTT	GCT CCA 620 GAT CTA 76	TGC ACG	75 ACC TGG GGC GGC	GCT CGA 763 AGC TCG	GAC CTG 0 TGA ACT	CCA CCT CTT CAA
7540 TAT GTG ATA CAC 7590 TTT GGA AAA CCT 7640 GTT GTG	75 TCT AGA AGA TCT TTC AAG	50 GAA CTT 760 CTT GAA 7	AAT TTA 0 AAG TTC 650 TAA ATT	CIC CIC CCIC	560 CTC GAG 76 GCG CCC	CCC CCC CCA CCT 766 CAC CTC	CAC CTC	757 CCC GCC 7 GAA CTT	GCT CCA 620 GAT CTA 76	TGC ACG	75 ACC TCC CCC CCC	GCT CGA 763 AGC TCG	GAC CTG 0 • TGA ACT 680 • CTG GAC	CCA CCT CTT CAA
7540 TAT GTG ATA CAC 7590 TTT GGA AAA CCT 7640 GTT GTG CAA CAC 769	75 TCT AGA AGA TCT TTC AAG	GAA CTT 760 CTT GAA 7 TGA ACT	AAT TTA 0 AAG TTC 650 TAA ATT 77	CTC  CAG  CTC  CAG  CTC	560 CTC CAG 76 CCG CCC	CCC CCC 10 CCA CCT 766 CAC CTC	CAC CTC CAA CTT O GTA CAT	757 CCC GCC GCC ACT TCA GTA	GCT CCA 620 GAT CTA 76 CCC CCC	TGC ACG	ACC TOG GGC CGC	GCT CGA 763 AGC TCG	GAC CTG 0 TGA ACT 680 CTG GAC	GCX CGT GTT CAA TTA AAT
7540 TAT GTG ATA CAC 7590 TTT GGA AAA CCT 7640 GTT GTG CAA CAC 769 ACG GTG TCC CAC	75 TCT AGA AGA TCT TTC AAG	GAA CTT 760 CTT GAA 7 TGA ACT	AAT TTA 0 AAG TTC 650 TAA ATT 77	CTC  CAG  CTC  CAG  CTC	TCA AGT	CCC CCC 10 CCA CCT 766 CAC CTC	CAC CTT CAT T10 CCA CCT	757 CCC GCC GCC ACT TCA GTA	GCT CCA 620 GAT CTA 76 CCC CCC	TGC ACG GCA CGT 70 GTT CAA 772 GTT CAA	ACC TOG GGC CGC	GCT CGA 763 AGC TCG	GAC CTG 0 TGA ACT 680 CTG GAC	GCX CGT GTT CAA TTA AAT
7540  TAT GTG ATA CAC  7590  TTT GGA AAA CCT  7640  GTT GTG CAA CAC  769  ACG GTG TGC CAC	75 TCT AGA AGA TCT TTC AAG GAG CTC	SO CAA CTT CAA 7 TCA ACT CCC	AAT TTA 0 AAG TTC 650 TAA ATT 77 AGT TCA	GAG CTC GAG CTC GTA CTA CAT	76 CCC CCC TCA ACT	CCCC 110 CCCA CCCT 7666 CACCCTCC 7 TCAA ACT	CAG CTC CAA CTT 0 CAT 710 CCA CCT	757 CCC GCC 7 GAA CTT TCA GTA CAT	GCT CCA 620 GAT CTA 76 CCC CCC	CCA CCT 70 CTT CAA	ACC CCC CCC CCC CCC CCC CCC CCC CCC CCC	FROM THE STATE OF	CAC CTG 0 + TGA ACT CTG GAC 77 CCG GAC	GCA CGT GTT CAA TTA AAT
7540 TAT GTG ATA CAC 7590 TTT GGA AAA CCT 7640 GTT GTG CAA CAC 769 ACG GTG TCC CAC	75 TCT AGA AGA TCT TTC AAG CTC 7740	SO CAT	AAT TTA  O AAG TTC  650 TAA ATT  77 AGT TCA	GAG CTC GAG CTC GTA CAT 775	76 CCC CAG CCC TCA ACT	CCCC  10  CCA  CCT  766  CAC  CTC  TGA  ACT	CAG CTC CAA CTT 0 GTA CAT 710 GCA CGT	757 CCCC GCC 7 GAA CTT TCA GTA CAT	GCT CCA 620 GAT CTA 76 CCC GCC	TCC ACC GCA CCT 70 GTT CAA GTT CAA	ACC CCC CCC CCC CCC CCC CCC CCC CCC CCC	FROM THE SECOND	CAC CTG 0 TGA ACT CTG GAC 77 CCG	GCA CGT GTT CAA TTA AAT
7540  TAT GTG ATA CAC  7590  TTT GGA AAA CCT  7640  GTT GTG CAA CAC  769  ACG GTG TGC CAC	75 TCT AGA AGA TCT TTC AAG CTC 7740 AGA TCT	SO CAT	AAT TTA  O AAG TTC  650 TAA ATT  77 AGT TCA	GAG CTC GCA CCT GAG CTA CAT 775 AGC	76 CCC CAG CCC TCA ACT	CCCC  10  CCA  CCT  766  CAC  CTC  TGA  ACT	CAG CTC CAA CTT 0 GTA CAT 710 GCA CGT	757 CCC GCC 7 GAA CTT TCA GTA CAT 760 AAC TTG	GCT CCA 620 GAT CTA 76 CCC GCC	TCC ACC GCA CCT 70 GTT CAA GTT CAA	ACC CCC CCC CCC CCC CCC CCC CCC CCC CCC	FROM THE SECOND	CAC CTG 0 TGA ACT CTG GAC 77 CCG	GCA CGT GTT CAA TTA AAT
7540 TAT GTG ATA CAC 7590 TTT GGA AAA CCT 7640 GTT GTG CAA CAC 769 ACG GTG TCC CAC	75 TCT AGA AGA TCT TTC AAG CTC 740 AGA TCT 77	TOA ACT	AAT TTA 0 AAG TTC 650 TAA AAT TCA	CAC CAT CAT CAT ACC TOS	76 GCC TCA TCA TCA TCA TCA TCA	CCCC 110 CCCT 766 CAG CCTC TCA ACT CAG CCTC	CAG CTC CAT O GTA CAT 710 GCA CGT 77 ACT TGA	757 CCC GCC GCC 7 GAA CCTT ACT TCA GTA CAT CAT 760 AAC TTC 78:	GCT CCA GAT CTA 76 CCC GCG	TGC AAG GCA CGT 70 GTT CAA 772 GTT CAA	ACC TTGG GGC CCG GCC GCC TTGC TTC AAG	FOR COLC COLC COLC COLC COLC COLC COLC CO	GAC CTG 0 TGA ACT 680 CTG GAC TTCC AGG	GCA CGT CAA TTA AAT CGC GCC

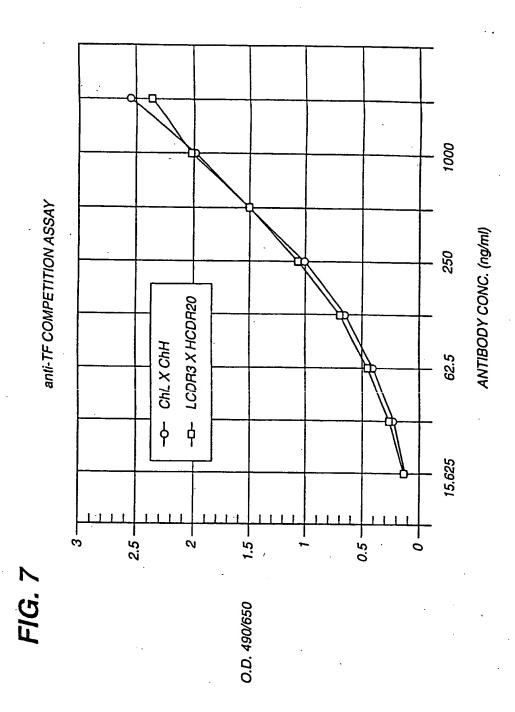
## FIG. 5 0

7830 7840 7850 7860

CGA TCG ACT CTA GAG GAT CGA TCC CCG GGC GAG CTC G
GCT AGC TGA GAT CTC CTA GCT AGG GGC CCG CTC GAG C

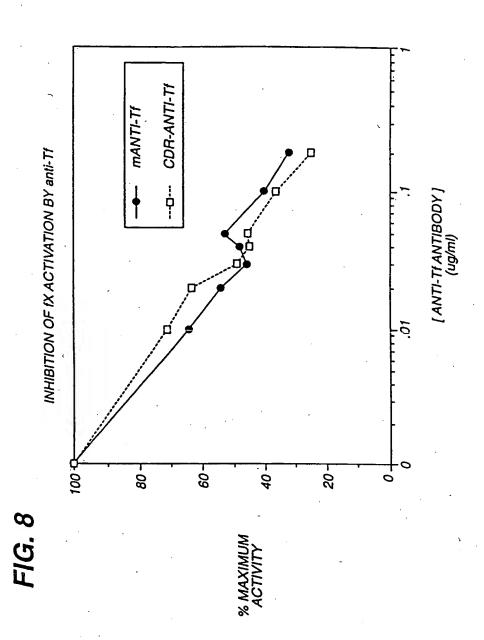


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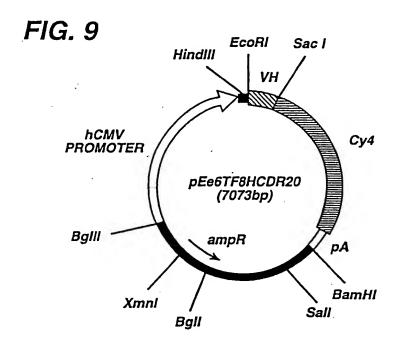


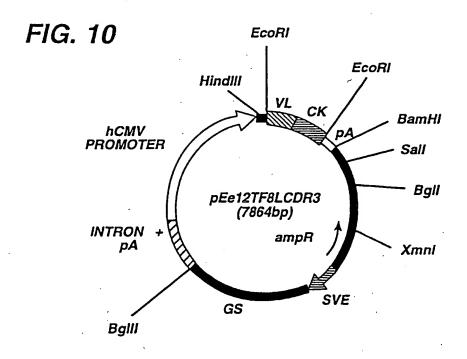
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onal Application No

PCT/US 96/09287 A. CLASSIFICATION OF SUBJECT MATTER
IPC 6 C12N15/13 C07K16/36 C07K16/46 A61K39/395 //C12N5/10, C12N15/85 According to International Patent Classification (IPC) or to both national classification and IPC **B. FIELDS SEARCHED** Minimum documentation searched (classification system followed by classification symbols) IPC 6 C12N C07K A61K Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched Electronic data base consulted during the international search (name of data base and, where practical, search terms used) C. DOCUMENTS CONSIDERED TO BE RELEVANT Citation of document, with indication, where appropriate, of the relevant passages Relevant to claim No. Category WO 91 09968 A (CELLTECH LIMITED) 11 July 1-37 1991 see examples see claims Υ WO 88 07543 A (SCRIPPS CLINIC AND RESEARCH 1-37 FOUNDATION) 6 October 1988 see claims WO 94 11029 A (THE SCRIPPS RESEARCH 1-37 INSTITUTE ET AL.) 26 May 1994 see claims WO 94 05328 A (THE SCRIPPS RESEARCH 1-37 INSTITUTE) 17 March 1994 see examples see claims -/--. Further documents are listed in the continuation of box C. X Patent family members are histed in annex. Special categories of cited documents: "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the 'A' document defining the general state of the art which is not considered to be of particular relevance "E" earlier document but published on or after the international "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone filing date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such docu-ments, such combination being obvious to a person skilled document referring to an oral disclosure, use, exhibition or document published prior to the international filing date but later than the priority date claimed "A" document member of the same patent family Date of the actual completion of the international search Date of mailing of the international search report 0 8. 11. 96 15 October 1996 Name and mailing address of the ISA Authorized officer

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Nooij, F

Intermal Application No PCI/US 96/09287

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Form PCT/ISA/218 (continuation of second sheet) (July 1992)

I national application No.

PCT/US 96/09287

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)
This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:
1. X Claims Nos.: 31-35 because they relate to subject matter not required to be searched by this Authority, namely: Remark: Although claims 31-35 are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.
2. Claims Nos.: because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
3. Claims Nos.: because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).
Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)
This International Searching Authority found multiple inventions in this international application, as follows:
As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
As all searchable claims could be searches without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims, it is covered by claims Nos.:
Remark on Protest  The additional search fees were accompanied by the applicant's protest.
No protest accompanied the payment of additional search fees.

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